

10/697,036

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FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 11:20:41 ON 02 OCT 2006

L1 86 S OSMOSENSING (2W) KINASE?  
L2 0 S "SACHAROMYCES(W) CERREVISIAE"  
L3 10 S SACCHAROMYCES(W) CERREVISIAE  
L4 0 S L1 AND L2  
L5 4046139 S HOST OR CLON? OR RECOMBINANT  
L6 25 S L1 AND L5  
L7 11 DUP REM L6 (14 DUPLICATES REMOVED)  
L8 456 S SLN1  
L9 43 S L5 AND L8  
L10 30 DUP REM L9 (13 DUPLICATES REMOVED)  
L11 0 S CRREVISIAE  
L12 11 S CERREVISIAE  
L13 0 S L10 AND L12  
E NAKAJIMA H/AU  
L14 8946 S E3  
L15 0 S L9 AND L14  
L16 1 S L1 AND L14

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=> s osmosensing (2w) kinase?  
L1 86 OSMOSENSING (2W) KINASE?

=> s "sacharomyces(w) cerevisiae"  
L2 0 "SACHAROMYCES(W) CERREVISIAE"

=> s saccharomyces(w) cerevisiae  
L3 10 SACCHAROMYCES(W) CERREVISIAE

=> s l1 and l2  
L4 0 L1 AND L2

=> s host or clon? or recombinant  
L5 4046139 HOST OR CLON? OR RECOMBINANT

=> s l1 and l5  
L6 25 L1 AND L5

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L7 11 DUP REM L6 (14 DUPLICATES REMOVED)

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L7 ANSWER 1 OF 11 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN  
DUPLICATE 1

ACCESSION NUMBER: 2006:250002 BIOSIS

DOCUMENT NUMBER: PREV200600250028

TITLE: Survey of mutations of a histidine kinase gene BcOS1 in  
dicarboximide-resistant field isolates of Botrytis cinerea.

AUTHOR(S): Oshima, Michiyo; Banno, Shinpei; Okada, Kiyotsugu;  
Takeuchi, Taeko; Kimura, Makoto; Ichiishi, Akihiko;  
Yamaguchi, Isamu; Fujimura, Makoto [Reprint Author]

CORPORATE SOURCE: Toyo Univ, Fac Life Sci, Gunma 3740193, Japan  
fujmura@itakura.toyo.ac.jp

SOURCE: Journal of General Plant Pathology, (FEB 2006) Vol. 72, No.  
1, pp. 65-73.  
ISSN: 1345-2630.

DOCUMENT TYPE: Article

LANGUAGE: English

ENTRY DATE: Entered STN: 26 Apr 2006

Last Updated on STN: 26 Apr 2006

AB Previously, we cloned a putative osmosensing histidine  
kinase gene (BcOS1) and revealed that a single amino acid  
substitution, isoleucine to serine at codon 365, conferred dicarboximide  
resistance in field isolates of Botrytis cinerea. This point mutation  
(type I) occurred within the restriction enzyme TaqI site of the wild-type  
BcOS1 gene. Thus, a procedure was developed for detecting the type I  
mutation of the BcOS1 gene using a polymerase chain reaction (PCR) in  
combination with restriction fragment-length polymorphism (RFLP).  
Diagnosis by PCR-RFLP was conducted on the 105 isolates isolated from 26  
fields in Japan. All dicarboximide-sensitive isolates (49 isolates) had  
the wild-type BcOS1 gene, and the 43 isolates with the type I mutation  
were resistant to dicarboximides without exception. These data indicate  
that dicarboximide-resistant isolates with type I mutation are widespread  
throughout Japan. However, other types of dicarboximide resistance were  
detected among isolates from Osaka; among the 24 resistant isolates from  
Osaka, 12 had the BcOS1 gene without the type I mutation. BcOS1 gene  
sequencing of these resistant isolates classified them into two groups,  
type II and type III. The type II isolates have three amino acid  
substitutions within BcOS1p ((368)Val to Phe, (369)Gln to His, and  
(447)Thr to Ser). The type III isolates have two amino acid substitutions  
within BcOS1p ((369)Gln to Pro and (373)Asn to Ser). These amino acid  
changes are located on the amino acid repeat domain in BcOS1p. The three  
types of resistant isolates were all moderately resistant to  
dicarboximides without significant osmotic sensitivity, and their  
pathogenicity on cucumber leaves was also very similar to that of the  
wild-type isolate.

L7 ANSWER 2 OF 11 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN  
DUPLICATE 2

ACCESSION NUMBER: 2004-15129 BIOTECHDS

TITLE: New transformed cell in which a polynucleotide coding for  
osmosensing histidine kinase having no  
transmembrane region has been introduced, useful for  
identifying an antifungal compound useful for killing a  
fungus;  
vector expression in host cell for use in drug  
screening and fungus infection therapy

AUTHOR: NAKAJIMA H

PATENT ASSIGNEE: SUMITOMO CHEM CO LTD

PATENT INFO: EP 1415996 6 May 2004

APPLICATION INFO: EP 2003-256895 30 Oct 2003

PRIORITY INFO: JP 2002-317736 31 Oct 2002; JP 2002-317736 31 Oct 2002  
DOCUMENT TYPE: Patent  
LANGUAGE: English  
OTHER SOURCE: WPI: 2004-341880 [32]

AB DERWENT ABSTRACT:

NOVELTY - A transformed cell in which a polynucleotide having a sequence encoding an amino acid sequence of an osmosensing histidine kinase having no transmembrane region has been introduced in a functional form into a cell deficient in at least one hybrid-sensor kinase, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for: (1) assaying the antifungal activity of a substance; (2) identifying an antifungal compound; (3) an antifungal compound selected by the method above; (4) killing a fungus; (5) an osmosensing histidine kinase having no transmembrane region and derived from a plant-pathogenic filamentous fungus or which has an amino acid sequence selected from: (a) a sequence of 1293, 1307 or 1438 amino acids (SEQ ID NO: 41, 55 or 68) given in the specification or a sequence 95% or more homologous to SEQ ID NO: 41, 55 or 68; (b) a sequence encoded by a DNA amplified by PCR using a *Fusarium oxysporum*-derived cDNA as a template and using oligonucleotides having the nucleotide sequences tgcactagtagtggttgacgacgcgccctcgc (SEQ ID NO: 52) and gagctgcagtttagttggaagacttcgcatatc (SEQ ID NO: 53) as primers; (c) a sequence encoded by a DNA amplified by PCR using *Mycosphaerella tritici*-derived cDNA as a template and using oligonucleotides having the sequences ccactagtagtgctgcaagaagagacttcg (SEQ ID NO: 64) and cctaagcttctcagctgctatgggcacgaa (SEQ ID NO: 65) as primers; (d) a sequence encoded by a DNA amplified by PCR using *Thanapethorus cucumeris*-derived cDNA as a template and using oligonucleotides having the sequences ggaactagtagtggtgacggtacaacggggggacacc (SEQ ID NO: 85) and tgcaagcttttagtggtgacggtggtgttacg (SEQ ID NO: 86) as primers; and (e) a sequence derived from *Phytophthora infestans* and has the amino acid sequence of 124 amino acids (SEQ ID NO: 90) given in the specification; (6) a polynucleotide having a nucleotide sequence encoding an amino acid sequence of an osmosensing histidine kinase having no transmembrane region derived from a plant-pathogen filamentous fungus described above or having a sequence of 3882, 3924, or 4317 bp (SEQ ID NO: 42, 56, or 69) given in the specification; (7) obtaining the polynucleotide above; and (8) an oligonucleotide which comprises a nucleotide sequence selected from 17 sequences of 23-34 bp (SEQ ID NO: 30-40, 52, 53, 64, 65, 85, and 86) given in the specification, e.g., aacatgtcccacgarattcgmacacc (SEQ ID NO: 30) caccgagattcgvacacccatgaaygg (SEQ ID NO: 31) aggccttcacaaaggctctvcggga (SEQ ID NO: 32) gagatggaccctgaaatcacmac (SEQ ID NO: 33) cagatattctcyagygaagtytckcg (SEQ ID NO: 34) atagcrttgccaacmaggttmagaataa (SEQ ID NO: 35) aacttgatggcrttkccaacmaggtt (SEQ ID NO: 36) ctctgtgaacttgatrgcrttkccaac (SEQ ID NO: 37) atacacttttcncgggtcacccatcat (SEQ ID NO: 38) tccatctgbgcctggatacacttttc (SEQ ID NO: 39) ggcttgavagataactcgtccatctg (SEQ ID NO: 40).

BIOTECHNOLOGY - Preferred Transformed Cell: The polynucleotide is a polynucleotide complementing the deficiency in hybrid-sensor kinase in the cell deficient in at least one hybrid-sensor kinase in which the polynucleotide has been introduced. The cell is a microorganism, particularly budding yeast. The osmosensing histidine kinase having no transmembrane region is an osmosensing histidine kinase having no transmembrane region and having a mutation which confers resistance to any of a dicarboxylimide antifungal compound, an aromatic hydrocarbon antifungal compound and a phenylpyrrole antifungal compound to the cell. The osmosensing histidine kinase is derived from a plant-pathogenic filamentous fungus and has no transmembrane region. The osmosensing histidine kinase has an amino acid sequence of 1315, 1307, 1293, 1307, 1438 or 124 amino acids (SEQ ID NO: 1, 16, 41, 55, 68 or 90, respectively) given in the specification. The nucleotide sequence encoding an amino

acid sequence of the osmosensing histidine kinase is a sequence of 3948, 3924, 3882, 3924, or 4317 bp (SEQ ID NO: 2, 17, 42, 56, or 69, respectively) also given in the specification. Preferred Method: Assaying the antifungal activity of a substance comprises culturing a transformed cell defined above in the presence of a test substance, measuring an amount of intracellular signal transduction from the osmosensing histidine kinase having no transmembrane region expressed in the cultured transformed cell or an index value having the correlation, and assessing the antifungal activity of the test substance based on a difference between an amount of intracellular signal transduction or an index value having the correlation measured and a control. The amount of intracellular signal transduction or the index value having the correlation is an amount of growth of the transformed cell. Identifying an antifungal compound comprises selecting an antifungal compound based on the antifungal activity assessed in the assaying method defined above. Killing a fungus comprises identifying an antifungal compound by the method above and contacting the fungus with the identified antifungal compound. Obtaining the polynucleotide above comprises amplifying a desired polynucleotide by PCR using the oligonucleotide above and recovering the amplified desired polynucleotide.

ACTIVITY - Fungicide. No biological data given.

MECHANISM OF ACTION - None given.

USE - The transformed cell is useful for assaying the antifungal activity of a substance and identifying an antifungal compound which is useful for killing a fungus (claimed).

EXAMPLE - BcOS-1 DNA was cloned into a shuttle vector p415ADH (ATCC87312) replicable in yeast and *Escherichia coli*. Both were digested, separated by agarose gel electrophoresis, and a part of the gel containing a desired DNA was excised. The BcOS-1 DNA was inserted between *Spe*I and *Pst*I sites in the multicloning site of the shuttle vector. A nucleotide sequence of the resulting expression plasmid was analyzed after a sequencing reaction. The nucleotide sequence of 3948 bp (SEQ ID NO: 2) given in the specification was obtained and it was confirmed that the expression plasmid pADHBcOS1 harbored a DNA having a nucleotide sequence encoding an amino acid sequence of BcOS-1. The prepared expression plasmid was introduced into each of budding yeast (*Saccharomyces cerevisiae* AH22 strain) (IFO10144) and TM182 strain. By utilizing disappearance of leucine auxotrophy in the resulting transformed budding yeast, the transformed budding yeast AH22 strain was selected on a Glu-Leu agar medium, and the transformed budding yeast TM182 was selected on a Gal-Ura-Leu agar medium. It was confirmed that the resulting TM182-BcOS1 grows even when transplanted to a Glu-Ura-Leu medium. (211 pages)

L7 ANSWER 3 OF 11 MEDLINE on STN  
ACCESSION NUMBER: 2003280893 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 12672798  
TITLE: Analysis of the role of the EnvZ linker region in signal transduction using a chimeric Tar/EnvZ receptor protein, Tez1.  
AUTHOR: Zhu Yan; Inouye Masayori  
CORPORATE SOURCE: Department of Biochemistry, Robert Wood Johnson Medical School, Piscataway, New Jersey 08854, USA.  
CONTRACT NUMBER: GM19043 (NIGMS)  
SOURCE: The Journal of biological chemistry, (2003 Jun 20) Vol. 278, No. 25, pp. 22812-9. Electronic Publication: 2003-04-02.  
Journal code: 2985121R. ISSN: 0021-9258.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200308

ENTRY DATE: Entered STN: 17 Jun 2003  
Last Updated on STN: 22 Aug 2003  
Entered Medline: 21 Aug 2003

AB Tez1 is a chimeric protein in which the periplasmic and transmembrane domains of Tar, a chemosensor, are fused to the cytoplasmic catalytic domain of EnvZ, an osmosensing histidine kinase, through the EnvZ linker. Unlike Taz1 (a similar hybrid with the Tar linker), Tez1 could not respond to Tar ligand, aspartate, whereas single Ala insertion at the transmembrane/linker junction, as seen in Tez1A1, restored the aspartate-regulatable phenotype. Analysis of the Ala insertion site requirement and the nature of the insertion residue on the phenotype of Tez1 indicated that a junction region between the transmembrane domain and the predicted helix I in the linker is critical to signal transduction. Random mutagenesis revealed that P185Q mutation in the Tez1 linker restored the aspartate-regulatable phenotype. Substitution mutations at Pro-185 further demonstrated that specific residues are required at this site for an aspartate response. None of the hybrid receptors constructed with different Tar/EnvZ fusion sites in the linker could respond to aspartate, suggesting that specific interactions between the two predicted helices in the linker are important for the linker function. In addition, a mutation (F220D) known to cause an OmpCc phenotype in EnvZ resulted in similar OmpCc phenotypes in both Tez1A1 and Tez1, indicating the importance of the predicted helix II in signal propagation. Together, we propose that the N-terminal junction region modulates the alignment between the two helices in the linker upon signal input. In turn helix II propagates the resultant conformational signal into the downstream catalytic domain of EnvZ to regulate its bifunctional enzymatic activities.

L7 ANSWER 4 OF 11 MEDLINE on STN DUPLICATE 3  
ACCESSION NUMBER: 2002387502 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 12135574  
TITLE: An osmosensing histidine kinase mediates dicarboximide fungicide resistance in *Botryotinia fuckeliana* (*Botrytis cinerea*).  
AUTHOR: Cui Wei; Beever Ross E; Parkes Stephanie L; Weeds Pauline L; Templeton Matthew D  
CORPORATE SOURCE: Plant Health and Development Group, The Horticulture and Food Research Institute of New Zealand Ltd., Private Bag 92 169, Auckland, New Zealand.  
SOURCE: Fungal genetics and biology : FG & B, (2002 Aug) Vol. 36, No. 3, pp. 187-98.  
Journal code: 9607601. ISSN: 1087-1845.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200210  
ENTRY DATE: Entered STN: 24 Jul 2002  
Last Updated on STN: 10 Oct 2002  
Entered Medline: 9 Oct 2002

AB A two-component histidine protein kinase gene, homologous to os-1 from *Neurospora crassa*, was cloned and sequenced from a single ascospore isolate of *Botryotinia fuckeliana*. A series of nine spontaneous mutants resistant to dicarboximide fungicides was selected from this strain and characterized with respect to fungicide resistance and osmotic sensitivity. Genetic crosses of the mutants with an authentic Dafi1 strain showed that the phenotypes mapped to this locus. Single point mutations (seven transitions, one transversion, and one short deletion) were detected in the alleles of the nine mutants sequenced. The mutational changes were shown to cosegregate with the dicarboximide resistance and osmotic sensitivity phenotypes in progeny obtained from crossing selected resistant strains with a sensitive strain. All mutations detected are predicted to result in amino acid changes in the coiled-coil region of the

putative Daf1 histidine kinase, and it is proposed that dicarboximide fungicides target this domain.

L7 ANSWER 5 OF 11 HCAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:12278 HCAPLUS  
DOCUMENT NUMBER: 137:89194  
TITLE: DNA fingerprint probe from *Mycosphaerella graminicola* identifies an active transposable element  
AUTHOR(S): Goodwin, Stephen B.; Cavaletto, Jessica R.; Waalwijk, Cees; Kema, Gert H. J.  
CORPORATE SOURCE: Crop Production and Pest Control Research, Purdue University, West Lafayette, IN, 47907, USA  
SOURCE: Phytopathology (2001), 91(12), 1181-1188  
CODEN: PHYTAJ; ISSN: 0031-949X  
PUBLISHER: American Phytopathological Society  
DOCUMENT TYPE: Journal  
LANGUAGE: English

AB DNA fingerprinting has been used extensively to characterize populations of *Mycosphaerella graminicola*, the *Septoria tritici* blotch pathogen of wheat. The highly polymorphic DNA fingerprints of *Mycosphaerella graminicola* were assumed to reflect the action of transposable elements. However, there was no direct evidence to support that conclusion. To test the transposable element hypothesis, the DNA fingerprint probe pSTL70 was sequenced, along with three other clones from a subgenomic library that hybridized with pSTL70. Anal. of these sequences revealed that pSTL70 contains the 3' end of a reverse transcriptase sequence plus 29- and 79-bp direct repeats. These are characteristics of transposable elements identified in other organisms. Southern analyses indicated that either the direct-repeat or reverse-transcriptase sequences by themselves essentially duplicated the original DNA fingerprint pattern, but other portions of pSTL70 contained single-copy DNA. Anal. of 60 progeny from a sexual cross between two Dutch isolates of *M. graminicola* identified several new bands that were not present in the parents. Thus, the putative transposable element probably is active during meiosis. Tests of single-spore isolates revealed gains or losses of one or more DNA fingerprint bands in 4 out of 10 asexual lines derived from isolate IPO94269. Therefore, DNA fingerprint patterns produced by the putative transposable element were capable of changes during asexual reproduction of this isolate. Probe pSTL70 did not hybridize at high stringency to genomic DNAs from other fungi related to *Septoria* and *Mycosphaerella*. These results indicate that the DNA fingerprint probe pSTL70 most likely identifies a transposable element in *M. graminicola* that may have been acquired recently, and appears to be active during both sexual and asexual reproduction

REFERENCE COUNT: 34 THERE ARE 34 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 6 OF 11 MEDLINE on STN

ACCESSION NUMBER: 2000223645 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 10760160  
TITLE: A monomeric histidine kinase derived from EnvZ, an *Escherichia coli* osmosensor.  
AUTHOR: Qin L; Dutta R; Kurokawa H; Ikura M; Inouye M  
CORPORATE SOURCE: Department of Biochemistry, UMDNJ, Robert Wood Johnson Medical School, 675 Hoes Lane, Piscataway, NJ 08854, USA.  
SOURCE: Molecular microbiology, (2000 Apr) Vol. 36, No. 1, pp. 24-32.  
Journal code: 8712028. ISSN: 0950-382X.  
PUB. COUNTRY: ENGLAND: United Kingdom  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200005  
ENTRY DATE: Entered STN: 6 Jun 2000



Last Updated on STN: 6 Jun 2000

Entered Medline: 23 May 2000

AB Histidine kinases function as dimers. The kinase domain of the osmosensing histidine kinase EnvZ of *Escherichia coli* consists of two domains: domain A (67 residues) responsible for histidine phosphotransfer and dimerization, and domain B (161 residues) responsible for the catalytic and ATP-binding function. The individual structures of these two domains have been recently solved by NMR spectroscopy. Here, we demonstrate that an enzymatically functional monomeric histidine kinase can be constructed by fusing in tandem two domains A and one domain B to produce a single polypeptide (A-A-B). We show that this protein, EnvZc[AAB], is soluble and exists as a stable monomer. The autophosphorylation and OmpR kinase activities of the monomeric EnvZc[AAB] are similar to that of the wild-type EnvZ, while OmpR-binding and phosphatase functions are reduced. V8 protease digestion and mutational analyses indicate that His-243 of only the amino proximal domain A is phosphorylated. Based on these results, molecular models are proposed for the structures of EnvZc[AAB] and the kinase domain of EnvZ. The present results demonstrate for the first time the construction of a functional, monomeric histidine kinase, further structural studies of which may provide important insights into the structure-function relationships of histidine kinases.

L7 ANSWER 7 OF 11 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN

DUPLICATE 4

ACCESSION NUMBER: 1999-13321 BIOTECHDS

TITLE: Osmosensing histidine-kinases, useful for screening for fungicide compounds and for the development of fungicide compounds;

*Candida albicans*, *Neurospora crassa* and *Saccharomyces cerevisiae* recombinant osmosensing thymidine-kinase, useful for drug screening for fungicide

AUTHOR: Alex L A; Simon M I; Selitrennikoff C; Agnan J

PATENT ASSIGNEE: Univ.Technol.Corp.; California-Inst.Technol.

LOCATION: Boulder, CO, USA; CA, USA.

PATENT INFO: US 5939306 17 Aug 1999

APPLICATION INFO: US 1997-843530 16 Apr 1997

PRIORITY INFO: US 1997-843530 16 Apr 1997

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 1999-468407 [39]

AB A purified and isolated DNA sequence (I) encoding an osmosensing histidine-kinase (II) (EC-2.7.1.21), along with vectors and host cells harboring (I) and its variants are new. (I) Is from *Candida albicans*, *Neurospora crassa* or *Saccharomyces cerevisiae*, and the preferred host cell for the production of (II) is *C. albicans*. The histidine-kinase is useful in drug screening for fungicides as well as for fungicide drug development. In an example, *N. crassa* was transfected with plasmid pMMS100 which was found to encode (II). (110pp)

L7 ANSWER 8 OF 11 MEDLINE on STN

DUPLICATE 5

ACCESSION NUMBER: 1998154430 MEDLINE

DOCUMENT NUMBER: PubMed ID: 9493379

TITLE: Isolation of CaSLN1 and CaNIK1, the genes for osmosensing histidine kinase homologues, from the pathogenic fungus *Candida albicans*.

AUTHOR: Nagahashi S; Mio T; Ono N; Yamada-Okabe T; Arisawa M; Bussey H; Yamada-Okabe H

CORPORATE SOURCE: Department of Biology, McGill University, Montreal, Quebec, Canada.

SOURCE: Microbiology (Reading, England), (1998 Feb) Vol. 144 ( Pt 2), pp. 425-32.

Journal code: 9430468. ISSN: 1350-0872.

PUB. COUNTRY: ENGLAND: United Kingdom  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
OTHER SOURCE: GENBANK-AB006362; GENBANK-AB006363  
ENTRY MONTH: 199804  
ENTRY DATE: Entered STN: 30 Apr 1998  
Last Updated on STN: 11 Feb 2003  
Entered Medline: 21 Apr 1998

AB Recent studies have revealed that fungi possess a mechanism similar to bacterial two-component systems to respond to extracellular changes in osmolarity. In *Saccharomyces cerevisiae*, Sln1p contains both histidine kinase and receiver (response regulator) domains and acts as an osmosensor protein that regulates the downstream HOG1 MAP kinase cascade. SLN1 of *Candida albicans* was functionally cloned using an *S. cerevisiae* strain in which SLN1 expression was conditionally suppressed. Deletion analysis of the cloned gene demonstrated that the receiver domain of *C. albicans* Sln1p was not necessary to rescue SLN1-deficient *S. cerevisiae* strains. Unlike *S. cerevisiae*, a null mutation of *C. albicans* SLN1 was viable under regular and high osmotic conditions, but it caused a slight growth retardation at high osmolarity. Southern blotting with *C. albicans* SLN1 revealed the presence of related genes, one of which is highly homologous to the NIK1 gene of *Neurospora crassa*. Thus, *C. albicans* harbours both SLN1- and NIK1-type histidine kinases.

L7 ANSWER 9 OF 11 MEDLINE on STN

ACCESSION NUMBER: 1999005252 MEDLINE

DOCUMENT NUMBER: PubMed ID: 9790591

TITLE: Yeast Skn7p activity is modulated by the Sln1p-Ypd1p osmosensor and contributes to regulation of the HOG pathway.

AUTHOR: Ketela T; Brown J L; Stewart R C; Bussey H

CORPORATE SOURCE: Department of Biology, McGill University, Montreal Quebec, Canada.

CONTRACT NUMBER: GM52583 (NIGMS)

SOURCE: Molecular & general genetics : MGG, (1998 Sep) Vol. 259, No. 4, pp. 372-8.

Journal code: 0125036. ISSN: 0026-8925.

PUB. COUNTRY: GERMANY: Germany, Federal Republic of

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199811

ENTRY DATE: Entered STN: 6 Jan 1999

Last Updated on STN: 11 Feb 2003

Entered Medline: 13 Nov 1998

AB Activation and control of the yeast HOG (High Osmolarity Glycerol) MAP kinase cascade is accomplished, in part, by a two-component sensory-response circuit comprised of the osmosensing histidine protein kinase Sln1p, the phospho-relay protein Ypd1p, and the response regulator protein Ssk1p. We found that deletion of SLN1 and/or YPD1 reduces reporter gene transcription driven by a second two-component response regulator -- Skn7p. The effect of sln1delta and ypd1delta mutations upon Skn7p activity is dependent on a functional two-component phosphorylation site (D427) in Skn7p, suggesting that Sln1p and Ypd1p may act as phosphodonors for Skn7p. We also observed that loss of PTC1 (a protein serine/threonine phosphatase implicated in negative control of the HOG pathway) in a skn7delta background results in severely retarded growth and in morphological defects. Deletion of either PBS2 or HOG1 alleviates the slow growth phenotype of ptc1delta skn7delta cells, suggesting that Skn7p may participate, in concert with known regulatory components, in modulating HOG pathway activity. The contribution of Skn7p to HOG pathway regulation appears to be modulated by the receiver domain, since non-phosphorylatable Skn7pD427N is unable to fully restore growth to

ptcl/skn7 cells.

L7 ANSWER 10 OF 11 MEDLINE on STN DUPLICATE 6  
ACCESSION NUMBER: 97287887 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 9142740  
TITLE: The osmotic-1 locus of *Neurospora crassa* encodes a putative histidine kinase similar to osmosensors of bacteria and yeast.  
AUTHOR: Schumacher M M; Enderlin C S; Selitrennikoff C P  
CORPORATE SOURCE: University of Colorado Health Sciences Center, B-111, 4200 East 9th Avenue, Denver, CO 80262, USA.  
CONTRACT NUMBER: R01:AI33354-02 (NIAID)  
SOURCE: Current microbiology, (1997 Jun) Vol. 34, No. 6, pp. 340-7. Journal code: 7808448. ISSN: 0343-8651.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Biotechnology  
OTHER SOURCE: GENBANK-U53189  
ENTRY MONTH: 199706  
ENTRY DATE: Entered STN: 16 Jul 1997  
Last Updated on STN: 16 Jul 1997  
Entered Medline: 30 Jun 1997

AB Osmotically sensitive mutants of *Neurospora crassa* are unable to grow on medium supplemented with 4% NaCl, have altered morphologies and cell-wall compositions, and are resistant to dicarboximide fungicides. Osmotic-1 (os-1) mutants have a unique characteristic of forming protoplasts that grow and divide in specialized liquid medium, suggesting that the os-1+ gene product is important for cell-wall assembly. A cosmid containing the os-1+ locus of *N. crassa*, isolated from a genomic cosmid library by chromosomal walk from a closely linked gene, was used to subclone the os-1+ gene by functional complementation of an os-1 mutant. Analysis of the sequence of complementing DNA predicts that os-1+ encodes a predicted protein similar to sensor-histidine kinases of bacteria and a yeast osmosensor-histidine kinase. Importantly, the predicted os-1+ protein is identical to the *N. crassa* nik-1 predicted protein that was identified by using polymerase chain reaction primers directed against histidine kinase consensus DNA sequences. Our results indicate that nik-1 and os-1 encode the same osmosensing histidine kinase that plays an important role in the regulation of cell-wall assembly and, probably, other cell responses to changes in external osmolarity.

L7 ANSWER 11 OF 11 LIFESCI COPYRIGHT 2006 CSA on STN  
ACCESSION NUMBER: 94:54454 LIFESCI  
TITLE: A two-component system that regulates an osmosensing MAP kinase cascade in yeast  
AUTHOR: Maeda, T.; Wurgler-Murphy, S.M.; Saito, H.\*  
CORPORATE SOURCE: Div. Tumor Immunol., Dana-Farber Cancer Inst., and Dep. Biol. Chem. and Mol. Pharmacol., Harvard Med. Sch., Boston, MA 02115, USA  
SOURCE: NATURE, (1994) vol. 369, no. 6477, pp. 242-245. ISSN: 0028-0836.  
DOCUMENT TYPE: Journal  
FILE SEGMENT: K; G  
LANGUAGE: English  
SUMMARY LANGUAGE: English

AB In the prokaryotic two-component signal transduction systems, recognition of an environmental stimulus by a sensor molecule results in the activation of its histidine kinase domain and phosphorylation of a histidine within that domain. This phosphate group is then transferred to an aspartate residue in the receiver domain of a cognate response regulator molecule, resulting in the activation of its output function. Although a few eukaryotic proteins were identified recently that show sequence similarity to the prokaryotic sensors or response regulators, it

has not been clear whether they constituted a part of a "two-component" system. Here we describe a two-component system in *Saccharomyces cerevisiae* that regulates an osmosensing MAP kinase cascade.

=> s sln1

L8 456 SLN1

=> d his

(FILE 'HOME' ENTERED AT 11:20:08 ON 02 OCT 2006)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 11:20:41 ON 02 OCT 2006

L1 86 S OSMOSENSING (2W) KINASE?  
L2 0 S "SACHAROMYCES(W) CERREVISIAE"  
L3 10 S SACCHAROMYCES(W) CERREVISIAE  
L4 0 S L1 AND L2  
L5 4046139 S HOST OR CLON? OR RECOMBINANT  
L6 25 S L1 AND L5  
L7 11 DUP REM L6 (14 DUPLICATES REMOVED)  
L8 456 S SLN1

=> s l5 and l8

L9 43 L5 AND L8

=> dup rem l9

PROCESSING COMPLETED FOR L9

L10 30 DUP REM L9 (13 DUPLICATES REMOVED)

=> s crrevisiae

L11 0 CRREVISIAE

=> s cerrevisiae

L12 11 CERREVISIAE

=> s l10 and l12

L13 0 L10 AND L12

=> d l10 1-30 ibib ab

L10 ANSWER 1 OF 30 HCAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2006:319146 HCAPLUS

DOCUMENT NUMBER: 144:366434

TITLE: Filamentation-defective variants of dimorphic fungi and their use in the treatment and prevention of candidiasis

INVENTOR(S): Milne, G. Todd

PATENT ASSIGNEE(S): Microbia, Inc., USA

SOURCE: PCT Int. Appl., 164 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2006036817	A2	20060406	WO 2005-US34219	20050923
WO 2006036817	A3	20060727		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KP, KR, KZ,

LC, LK, LR, LS, LT, LU, LV, LY, MA, MD, MG, MK, MN, MW, MX, MZ,  
 NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG,  
 SK, SL, SM, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN,  
 YU, ZA, ZM, ZW  
 RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE,  
 IS, IT, LT, LU, LV, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ,  
 CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG, BW, GH,  
 GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY,  
 KG, KZ, MD, RU, TJ, TM

PRIORITY APPLN. INFO.: US 2004-613112P P 20040924

AB Methods of inhibiting and treating infection by dimorphic fungi using strains of the fungus that cannot transition from the yeast habit to the filamentous growth habit are described. These strain can also be used to deliver antigens in vaccines by presenting them on the cell surface. A filamentation-incompetent strain of *Candida albicans* was constructed by deleting both copies of the *CRV1* gene. Mice inoculated with cells of this mutant were resistant to a subsequent challenge with a wild-type *C. albicans*.

L10 ANSWER 2 OF 30 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 2006:81117 LIFESCI

TITLE: A Downshift in Temperature Activates the High Osmolarity Glycerol (HOG) Pathway, Which Determines Freeze Tolerance in *Saccharomyces cerevisiae*

AUTHOR: Panadero, Joaquin; Pallotti, Claudia; Rodriguez-Vargas, Sonia; Randez-Gil, Francisca; Prieto, Jose A.

CORPORATE SOURCE: Department of Biotechnology, Instituto de Agroquimica y Tecnologia de los Alimentos, Consejo Superior de Investigaciones Cientificas, P. O. Box 73, E-46100-Burjassot Valencia, Spain

SOURCE: Journal of Biological Chemistry [J. Biol. Chem.], (20060200 vol. 281, no. 8, pp. 4638-4645. ISSN: 0021-9258.

DOCUMENT TYPE: Journal

FILE SEGMENT: K; N

LANGUAGE: English

SUMMARY LANGUAGE: English

AB The molecular mechanisms that enable yeast cells to detect and transmit cold signals and their physiological significance in the adaptive response to low temperatures are unknown. Here, we have demonstrated that the MAPK Hoglp is specifically activated in response to cold. Phosphorylation of Hoglp was dependent on Pbs2p, the MAPK kinase (MAPKK) of the high osmolarity glycerol (HOG) pathway, and Ssk1p, the response regulator of the two-component system Sln1p-Ypd1p. However, Sholp was not required. Interestingly, phosphorylation of Hoglp was stimulated at 30 degree C in cells exposed to the membrane rigidifier agent dimethyl sulfoxide. Moreover, Hoglp activation occurred specifically through the Sln1 branch. This suggests that Sln1 monitors changes in membrane fluidity caused by cold. Quite remarkably, activation of Hoglp at low temperatures affected the transcriptional response to cold shock. Indeed, the absence of Hoglp impaired the cold-instigated expression of genes for trehalose- and glycerol-synthesizing enzymes and small chaperones. Moreover, a downward transfer to 12 or 4 degree C stimulated the overproduction of glycerol in a Hoglp-dependent manner. However, hog1 Delta mutant cells showed no growth defects at 12 degree C as compared with the wild type. On the contrary, deletion of HOG1 or GPD1 decreased tolerance to freezing of wild-type cells preincubated at a low temperature, whereas no differences could be detected in cells shifted directly from 30 to -20 degree C. Thus, exposure to low temperatures triggered a Hoglp-dependent accumulation of glycerol, which is essential for freeze protection.

L10 ANSWER 3 OF 30 HCAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2006:385775 HCAPLUS

DOCUMENT NUMBER: 145:77422

TITLE: Global Control of Dimorphism and Virulence in Fungi  
 AUTHOR(S): Nemecek, Julie C.; Wuethrich, Marcel; Klein, Bruce S.  
 CORPORATE SOURCE: Department of Medical Microbiology and Immunology,  
 Univ. Wisconsin Hosp. and Clin., Madison, WI, 53792,  
 USA  
 SOURCE: Science (Washington, DC, United States) (2006),  
 312(5773), 583-588  
 CODEN: SCIEAS; ISSN: 0036-8075  
 PUBLISHER: American Association for the Advancement of Science  
 DOCUMENT TYPE: Journal  
 LANGUAGE: English  
 AB Microbial pathogens that normally inhabit our environment can adapt to  
 thrive inside mammalian hosts. There are six dimorphic fungi  
 that cause disease worldwide, which switch from nonpathogenic molds in  
 soil to pathogenic yeast after spores are inhaled and exposed to elevated  
 temperature. Mechanisms that regulate this switch remain obscure. We show that  
 a hybrid histidine kinase senses host signals and triggers the  
 transition from mold to yeast. The kinase also regulates cell-wall  
 integrity, sporulation, and expression of virulence genes in vivo. This  
 global regulator shapes how dimorphic fungal pathogens adapt to the  
 mammalian host, which has broad implications for treating and  
 preventing systemic fungal disease.  
 REFERENCE COUNT: 31 THERE ARE 31 CITED REFERENCES AVAILABLE FOR THIS  
 RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 4 OF 30 MEDLINE on STN  
 ACCESSION NUMBER: 2005078632 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 15707964  
 TITLE: Evidence that C-terminal non-kinase domain of Pbs2p has a  
 role in high osmolarity-induced nuclear localization of  
 Hog1p.  
 AUTHOR: Sharma Pratima; Mondal Alok K  
 CORPORATE SOURCE: Institute of Microbial Technology, Sector 39A, Chandigarh  
 160 036, India.  
 SOURCE: Biochemical and biophysical research communications, (2005  
 Mar 25) Vol. 328, No. 4, pp. 906-13.  
 Journal code: 0372516. ISSN: 0006-291X.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200504  
 ENTRY DATE: Entered STN: 16 Feb 2005  
 Last Updated on STN: 9 Apr 2005  
 Entered Medline: 8 Apr 2005  
 AB Mitogen-activated protein kinase (MAPK) cascade is a ubiquitous signaling  
 module that transmits extracellular stimuli through the cytoplasm to the  
 nucleus. In baker's yeast external high osmolarity activates high  
 osmolarity glycerol (HOG) MAPK pathway which consists of two upstream  
 branches (SHO1 and SLN1) and common downstream elements Pbs2p  
 MAPKK and Hog1p MAPK. Activation of this pathway causes rapid nuclear  
 accumulation of Hog1p, essentially leading to the expression of target  
 genes. Previously we have isolated a PBS2 homologue (DPBS2) from  
 osmo-tolerant and salt-tolerant yeast *Debaryomyces hansenii* that partially  
 complemented pbs2 mutation in *Saccharomyces cerevisiae*. Here we show that  
 by replacing C-terminal region of Dpbs2p with the homologous region of  
 Pbs2p we could abrogate partial complementation exhibited by Dpbs2p and  
 this was achieved due to increase in nuclear translocation of Hog1p.  
 Thus, our result showed that in HOG pathway, MAPKK has important role in  
 nuclear translocation of Hog1p.

L10 ANSWER 5 OF 30 MEDLINE on STN  
 ACCESSION NUMBER: 2005003693 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 15628880

TITLE: Kinetic analysis of YPD1-dependent phosphotransfer reactions in the yeast osmoregulatory phosphorelay system.  
 AUTHOR: Janiak-Spens Fabiola; Cook Paul F; West Ann H  
 CORPORATE SOURCE: Department of Chemistry and Biochemistry, University of Oklahoma, Norman, Oklahoma 73019, USA.  
 CONTRACT NUMBER: GM59311 (NIGMS)  
 SOURCE: Biochemistry, (2005 Jan 11) Vol. 44, No. 1, pp. 377-86.  
 Journal code: 0370623. ISSN: 0006-2960.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200503  
 ENTRY DATE: Entered STN: 5 Jan 2005  
 Last Updated on STN: 5 Mar 2005  
 Entered Medline: 4 Mar 2005

AB In *Saccharomyces cerevisiae*, the histidine-containing phosphotransfer (HPT) protein YPD1 transfers phosphoryl groups between the three different response regulator domains of SLN1, SSK1, and SKN7 (designated R1, R2, and R3, respectively). Together these proteins form a branched histidine-aspartic acid phosphorelay system through which cells can respond to hyperosmotic and other environmental stresses. The in vivo order of phosphotransfer reactions is believed to proceed from SLN1-R1 to YPD1 and then subsequently to SSK1-R2 or SKN7-R3. The individual phosphoryl transfer reactions between YPD1 and the response regulator domains have been examined kinetically. A maximum forward rate constant of 29 s<sup>-1</sup> was determined for the reaction between SLN1-R1 approximately P and YPD1 with a K<sub>d</sub> of 1.4 microm for the SLN1-R1 approximately P.YPD1 complex. In the subsequent reactions, phosphotransfer from YPD1 to SSK1-R2 is very rapid (160 s<sup>-1</sup>) and is strongly favored over phosphotransfer to SKN7-R3. Phosphotransfer reactions between YPD1 and SLN1-R1 or SKN7-R3 were reversible. In contrast, no reverse transfer from SSK1-R2 approximately P to YPD1 was observed. These findings are consistent with the notion that SSK1 is constitutively phosphorylated under normal osmotic conditions. In addition, we have examined the roles of several conserved amino acid residues surrounding the phosphorylatable histidine (H64) of YPD1 using phosphoryl transfer reactions involving YPD1 mutants. With respect to phosphoryl transfer from SLN1-R1 approximately P, only one YPD1 mutant (K67A) exhibited an increase in K<sub>d</sub> and thus affects binding of YPD1 to SLN1-R1 approximately P, whereas other mutants (R90A, Q86A, and G68Q) showed a decrease in phosphoryl transfer rate. Only the G68Q-YPD1 mutant was significantly affected in phosphotransfer to SSK1-R2 (approximately 680-fold decrease in rate in comparison to wild-type). This is the first report of a kinetic analysis of a eukaryotic "two-component" histidine-aspartic acid phosphotransfer system, enabling a comparison of the transfer rates and binding constants to the few bacterial systems that have been studied this way.

L10 ANSWER 6 OF 30 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN  
 ACCESSION NUMBER: 2005:169412 BIOSIS  
 DOCUMENT NUMBER: PREV200500170366  
 TITLE: Pradimicin resistance of yeast is caused by a mutation of the putative N-glycosylation sites of osmosensor protein Sln1.  
 AUTHOR(S): Hiramoto, Fumitaka; Nomura, Nobuhiko; Furumai, Tamotsu; Igarashi, Yasuhiro [Reprint Author]; Oki, Toshikazu  
 CORPORATE SOURCE: Biotechnol Res Ctr, Toyama Prefectural Univ, 5180 Kurokawa, Toyama, 9390398, Japan  
 yas@pu-toyama.ac.jp  
 SOURCE: Bioscience Biotechnology and Biochemistry, (January 2005)  
 Vol. 69, No. 1, pp. 238-241. print.  
 ISSN: 0916-8451.  
 DOCUMENT TYPE: Article

LANGUAGE: English  
ENTRY DATE: Entered STN: 4 May 2005  
Last Updated on STN: 4 May 2005

AB Pradimicin, a mannose-binding antifungal antibiotic, induces apoptosis-like cell death in *Saccharomyces cerevisiae*. Previously we found that the substitution of the 74th amino acid from glycine to cysteine in Ypd1 yields a mutant resistant to pradimicin. In this study, the involvement of a membrane-spanning osmosensor, Sln1, which is located upstream of Ypd1, was investigated. A mutant, sln1 ANG, that lacks the putative N-glycosylation sites in the extracellular domain became resistant to pradimicin. On the other hand, the null mutants of Ssk1, Pbs2, and Hog1, which are located downstream of the Sln1 cascade, were sensitive to pradimicin as well as the wild-type strain. In conclusion, pradimicin exerts its fungicidal action with the involvement of Sln1, but the downstream branch, Ssk1 and the HOG pathway, is not involved.

L10 ANSWER 7 OF 30 MEDLINE on STN  
ACCESSION NUMBER: 2004616803 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 15590828  
TITLE: Role for the Ran binding protein, Moglp, in *Saccharomyces cerevisiae* SLN1-SKN7 signal transduction.  
AUTHOR: Lu Jade Mei-Yeh; Deschenes Robert J; Fassler Jan S  
CORPORATE SOURCE: Department of Biological Sciences, University of Iowa, 202 BBE, Iowa City, IA 52242, USA.  
CONTRACT NUMBER: GM56719 (NIGMS)  
SOURCE: Eukaryotic cell, (2004 Dec) Vol. 3, No. 6, pp. 1544-56.  
Journal code: 101130731. ISSN: 1535-9778.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200506  
ENTRY DATE: Entered STN: 20 Dec 2004  
Last Updated on STN: 29 Jun 2005  
Entered Medline: 28 Jun 2005

AB Yeast Sln1p is an osmotic stress sensor with histidine kinase activity. Modulation of Sln1 kinase activity in response to changes in the osmotic environment regulates the activity of the osmotic response mitogen-activated protein kinase pathway and the activity of the Skn7p transcription factor, both important for adaptation to changing osmotic stress conditions. Many aspects of Sln1 function, such as how kinase activity is regulated to allow a rapid response to the continually changing osmotic environment, are not understood. To gain insight into Sln1p function, we conducted a two-hybrid screen to identify interactors. Moglp, a protein that interacts with the yeast Ran1 homolog, Gsplp, was identified in this screen. The interaction with Moglp was characterized in vitro, and its importance was assessed in vivo. mog1 mutants exhibit defects in SLN1-SKN7 signal transduction and mislocalization of the Skn7p transcription factor. The requirement for Moglp in normal localization of Skn7p to the nucleus does not fully account for the mog1-related defects in SLN1-SKN7 signal transduction, raising the possibility that Moglp may play a role in Skn7 binding and activation of osmotic response genes.

L10 ANSWER 8 OF 30 MEDLINE on STN  
ACCESSION NUMBER: 2003356464 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 12853477  
TITLE: A docking site determining specificity of Pbs2 MAPKK for Ssk2/Ssk22 MAPKKs in the yeast HOG pathway.  
AUTHOR: Tatebayashi Kazuo; Takekawa Mutsuhiro; Saito Haruo  
CORPORATE SOURCE: Division of Molecular Cell Signaling, Institute of Medical Science, The University of Tokyo, 4-6-1 Shirokanedai, Minato-ku, Tokyo 108-8639, Japan.



SOURCE: The EMBO journal, (2003 Jul 15) Vol. 22, No. 14, pp. 3624-34.  
Journal code: 8208664. ISSN: 0261-4189.

PUB. COUNTRY: England: United Kingdom

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200309

ENTRY DATE: Entered STN: 1 Aug 2003  
Last Updated on STN: 24 Sep 2003  
Entered Medline: 23 Sep 2003

AB Mitogen-activated protein kinase (MAPK) cascades are conserved signaling modules composed of three sequentially activated kinases (MAPKKK, MAPKK and MAPK). Because individual cells contain multiple MAPK cascades, mechanisms are required to ensure the fidelity of signal transmission. In yeast, external high osmolarity activates the HOG (high osmolarity glycerol) MAPK pathway, which consists of two upstream branches (SHO1 and SLN1) and common downstream elements including the Pbs2 MAPKK and the Hog1 MAPK. The Ssk2/Ssk22 MAPKKs in the SLN1 branch, when activated, exclusively phosphorylate the Pbs2 MAPKK. We found that this was due to an Ssk2/Ssk22-specific docking site in the Pbs2 N-terminal region. The Pbs2 docking site constitutively bound the Ssk2/Ssk22 kinase domain. Docking site mutations drastically reduced the Pbs2-Ssk2/Ssk22 interaction and hampered Hog1 activation by the SLN1 branch. Fusion of the Pbs2 docking site to a different MAPKK, Ste7, allowed phosphorylation of Ste7 by Ssk2/Ssk22. Thus, the docking site contributes to both the efficiency and specificity of signaling. During these analyses, we also found a nuclear export signal and a possible nuclear localization signal in Pbs2.

L10 ANSWER 9 OF 30 MEDLINE on STN

ACCESSION NUMBER: 2003584403 MEDLINE

DOCUMENT NUMBER: PubMed ID: 14665464

TITLE: *Saccharomyces cerevisiae* histidine phosphotransferase Ypd1p shuttles between the nucleus and cytoplasm for SLN1-dependent phosphorylation of Ssk1p and Skn7p.

AUTHOR: Lu Jade Mei-Yeh; Deschenes Robert J; Fassler Jan S

CORPORATE SOURCE: Departments of Biological Sciences and Biochemistry, University of Iowa, Iowa City, Iowa 52242, USA.

CONTRACT NUMBER: GM-56719 (NIGMS)

SOURCE: Eukaryotic cell, (2003 Dec) Vol. 2, No. 6, pp. 1304-14.  
Journal code: 101130731. ISSN: 1535-9778.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200407

ENTRY DATE: Entered STN: 16 Dec 2003  
Last Updated on STN: 1 Aug 2004  
Entered Medline: 30 Jul 2004

AB Sln1p is a plasma membrane-localized two-component histidine kinase that functions as an osmotic stress sensor in *Saccharomyces cerevisiae*. Changes in osmotic pressure modulate Sln1p kinase activity, which, together with Ypd1p, a phosphorelay intermediate, changes the phosphorylation status of two response regulators, Ssk1p and Skn7p. Ssk1p controls the activity of the HOG1 mitogen-activated protein kinase pathway. Skn7p is a nuclearly localized transcription factor that regulates genes involved in cell wall integrity and other processes. Subcellular compartmentalization may therefore play an important role in eukaryotic two-component pathway regulation. We have studied the subcellular localization of SLN1 pathway components and find that Ypd1p is a dynamic protein with a role in shuttling the osmotic stress signal from Sln1p to Ssk1p in the cytosol and to Skn7p in the nucleus. The need to translocate the signal into different intracellular

compartments contributes a spatial dimension to eukaryotic two-component pathways compared to the prototypical two-component pathways of prokaryotes.

L10 ANSWER 10 OF 30 MEDLINE on STN  
ACCESSION NUMBER: 2003072192 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 12581306  
TITLE: A role for HvGAMYB in anther development.  
AUTHOR: Murray Fiona; Kalla Roger; Jacobsen John; Gubler Frank  
CORPORATE SOURCE: CSIRO Plant Industry, GPO Box 1600, Canberra City, Australia.  
SOURCE: The Plant journal : for cell and molecular biology, (2003 Feb) Vol. 33, No. 3, pp. 481-91.  
Journal code: 9207397. ISSN: 0960-7412.  
PUB. COUNTRY: England: United Kingdom  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200304  
ENTRY DATE: Entered STN: 14 Feb 2003  
Last Updated on STN: 30 Apr 2003  
Entered Medline: 29 Apr 2003

AB HvGAMYB is a transcription factor that was first identified in barley aleurone cells and shown to be upregulated by gibberellin (GA). Using RNA and immunoblot analysis we have shown HvGAMYB is also strongly expressed in barley anthers. Transgenic barley expressing a HvGAMYB:GFP fusion gene have been created and GAMYB expression in anthers analysed. GFP expression was clearly visible during early anther development in the nuclei of the epidermis, endothecium, middle layer and tapetum. Expression in the epidermis and endothecium persists until just prior to anther dehiscence, expression in the other two cell layers is visible until they are compressed and broken down as the microspores develop. Further evidence of a role for HvGAMYB in anther development was provided by the creation of transgenic barley over-expressing the HvGAMYB gene. Associated with the increase in HvGAMYB levels was a progressive decrease in anther size, particularly a decrease in anther length. Anthers also became increasingly lighter in colour. Anthers with fourfold more HvGAMYB protein than non-transgenic controls failed to dehisce and were male sterile, anthers with approximately three to fourfold endogenous GAMYB protein levels were smaller and paler but still shed normally. To investigate the hormonal regulation of HvGAMYB expression in anthers, HvGAMYB and SLN1 protein levels in anthers were analysed following application of GA3. As in cereal aleurone, HvGAMYB levels were found to increase and SLN1 levels decrease following GA3 application suggesting a similar GA-signalling pathway to that in aleurone exists in anthers.

L10 ANSWER 11 OF 30 MEDLINE on STN DUPLICATE 1  
ACCESSION NUMBER: 2002662570 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 12406718  
TITLE: Isolation and functional analysis of a gene, tcsB, encoding a transmembrane hybrid-type histidine kinase from *Aspergillus nidulans*.  
AUTHOR: Furukawa Kentaro; Katsuno Yasuaki; Urao Takeshi; Yabe Tomio; Yamada-Okabe Toshiko; Yamada-Okabe Hisafumi; Yamagata Youhei; Abe Keietsu; Nakajima Tasuku  
CORPORATE SOURCE: Laboratory of Enzymology, Department of Molecular and Cell Biology, Graduate School of Agricultural Science, Tohoku University, 1-1 Amamiya, Tsutsumi-dori, Sendai 981-8555, Japan.  
SOURCE: Applied and environmental microbiology, (2002 Nov) Vol. 68, No. 11, pp. 5304-10.  
Journal code: 7605801. ISSN: 0099-2240.  
PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200212  
ENTRY DATE: Entered STN: 9 Nov 2002  
Last Updated on STN: 18 Dec 2002  
Entered Medline: 17 Dec 2002

AB We cloned and characterized a novel *Aspergillus nidulans* histidine kinase gene, *tcsB*, encoding a membrane-type two-component signaling protein homologous to the yeast osmosensor synthetic lethal N-end rule protein 1 (SLN1), which transmits signals through the high-osmolarity glycerol response 1 (HOG1) mitogen-activated protein kinase (MAPK) cascade in yeast cells in response to environmental osmotic stimuli. From an *A. nidulans* cDNA library, we isolated a positive clone containing a 3,210-bp open reading frame that encoded a putative protein consisting of 1,070 amino acids. The predicted *tcsB* protein (TcsB) has two probable transmembrane regions in its N-terminal half and has a high degree of structural similarity to yeast Sln1p, a transmembrane hybrid-type histidine kinase. Overexpression of the *tcsB* cDNA suppressed the lethality of a temperature-sensitive osmosensing-defective *sln1-ts* yeast mutant. However, *tcsB* cDNAs in which the conserved phosphorylation site His(552) residue or the phosphorelay site Asp(989) residue had been replaced failed to complement the *sln1-ts* mutant. In addition, introduction of the *tcsB* cDNA into an *sln1delta* *sholdelta* yeast double mutant, which lacked two osmosensors, suppressed lethality in high-salinity media and activated the HOG1 MAPK. These results imply that TcsB functions as an osmosensor histidine kinase. We constructed an *A. nidulans* strain lacking the *tcsB* gene (*tcsBdelta*) and examined its phenotype. However, unexpectedly, the *tcsBdelta* strain did not exhibit a detectable phenotype for either hyphal development or morphology on standard or stress media. Our results suggest that *A. nidulans* has more complex and robust osmoregulatory systems than the yeast SLN1-HOG1 MAPK cascade.

L10 ANSWER 12 OF 30 MEDLINE on STN

ACCESSION NUMBER: 2002731366 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 12492836  
TITLE: Evidence that the *Arabidopsis* nuclear gibberellin signalling protein GAI is not destabilised by gibberellin.  
AUTHOR: Fleck Barbara; Harberd Nicholas P  
CORPORATE SOURCE: John Innes Centre, Norwich Research Park, Colney Lane, Norwich NR4 7UJ, UK.  
SOURCE: The Plant journal : for cell and molecular biology, (2002 Dec) Vol. 32, No. 6, pp. 935-47.  
Journal code: 9207397. ISSN: 0960-7412.  
PUB. COUNTRY: England: United Kingdom  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200305  
ENTRY DATE: Entered STN: 21 Dec 2002  
Last Updated on STN: 6 May 2003  
Entered Medline: 5 May 2003

AB Plant growth is regulated by bioactive gibberellin (GA), although there is an unexplained diversity in the magnitude of the GA responses exhibited by different plant species. GA acts via a group of orthologous proteins known as the DELLA proteins. The *Arabidopsis* genome contains genes encoding five different DELLA proteins, the best known of which are GAI and RGA. The DELLA proteins are thought to act as repressors of GA-regulated processes, whilst GA is thought to act as a negative regulator of DELLA protein function. Recent experiments have shown that GA induces rapid disappearance of nuclear RGA, SLR1 and SLN1 (DELLA proteins from rice and barley), suggesting that GA signalling and degradation of DELLA proteins are coupled. However, RGL1, another

Arabidopsis DELLA protein, does not disappear from the nucleus in response to GA treatment. Here, we present evidence suggesting that GAI, like RGL1, is stable in response to GA treatment, and show that transgenic Arabidopsis plants containing constructs that enable high-level expression of GAI exhibit a dwarf, GA non-responsive phenotype. Thus, GAI appears to be less affected by GA than RGA, SLR1 or SLN1. We also show that neither of the two putative nuclear localisation signals contained in DELLA proteins are individually necessary for nuclear localisation of GAI. The various DELLA proteins have different properties, and we suggest that this functional diversity may explain, at least in part, why plant species differ widely in their GA response magnitudes.

L10 ANSWER 13 OF 30 MEDLINE on STN  
 ACCESSION NUMBER: 2002264870 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 11985722  
 TITLE: A cytoplasmic coiled-coil domain is required for histidine kinase activity of the yeast osmosensor, SLN1.  
 AUTHOR: Tao Wei; Malone Cheryl L; Ault Addison D; Deschenes Robert J; Fassler Jan S  
 CORPORATE SOURCE: Department of Biological Sciences, University of Iowa, Iowa City 52242, USA.  
 CONTRACT NUMBER: GM56719 (NIGMS)  
 T32AG (NIA)  
 SOURCE: Molecular microbiology, (2002 Jan) Vol. 43, No. 2, pp. 459-73.  
 Journal code: 8712028. ISSN: 0950-382X.  
 PUB. COUNTRY: England: United Kingdom  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200208  
 ENTRY DATE: Entered STN: 14 May 2002  
 Last Updated on STN: 11 Feb 2003  
 Entered Medline: 2 Aug 2002

AB The yeast histidine kinase, Sln1p, is a plasma membrane-associated osmosensor that regulates the activity of the osmotic stress MAP kinase pathway. Changes in the osmotic environment of the cell influence the autokinase activity of the cytoplasmic kinase domain of Sln1p. Neither the nature of the stimulus, the mechanism by which the osmotic signal is transduced nor the manner in which the kinase is regulated is currently clear. We have identified several mutations located in the linker region of the Sln1 kinase (just upstream of the kinase domain) that cause hyperactivity of the Sln1 kinase. This region of histidine kinases is largely uncharacterized, but its location between the transmembrane domains and the cytoplasmic kinase domain suggests that it may have a potential role in signal transduction. In this study, we have investigated the Sln1 linker region in order to understand its function in signal transduction and regulation of Sln1 kinase activity. Our results indicate that the linker region forms a coiled-coil structure and suggest a mechanism by which alterations induced by osmotic stress influence kinase activity by altering the alignment of the phospho-accepting histidine with respect to the catalytic domain of the kinase.

L10 ANSWER 14 OF 30 MEDLINE on STN  
 ACCESSION NUMBER: 2002272526 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 12011349  
 TITLE: Mutants at the Slender1 locus of barley cv Himalaya. Molecular and physiological characterization.  
 AUTHOR: Chandler Peter Michael; Marion-Poll Annie; Ellis Marc; Gubler Frank  
 CORPORATE SOURCE: Commonwealth Scientific and Industrial Research Organization, Plant Industry, G.P.O. Box 1600, Canberra, Australian Capitol Territory 2601, Australia..

SOURCE: peter.chandler@csiro.au  
Plant physiology, (2002 May) Vol. 129, No. 1, pp. 181-90.  
Journal code: 0401224. ISSN: 0032-0889.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 200208  
ENTRY DATE: Entered STN: 16 May 2002  
Last Updated on STN: 10 Aug 2002  
Entered Medline: 9 Aug 2002

AB A dominant dwarf mutant of barley (*Hordeum vulgare*) that resembles dominant gibberellin (GA) "-insensitive" or "-nonresponsive" mutants in other species is described. alpha-Amylase production by endosperm half-grains of the mutant required GA3 at concentrations about 100 times that of the WT. The mutant showed only a slight growth response to GA3, even at very high concentrations. However, when additionally dwarfed, growth rate responded to GA3 over the normal concentration range, although only back to the original (dwarf) elongation rate. Genetic studies indicated that the dominant dwarf locus was either closely linked or identical to the *Sln1* (Slender1) locus. A barley sequence related to Arabidopsis GAI/RGA was isolated, and shown to represent the *Sln1* locus by the analysis of *sln1* mutants. The dominant dwarf mutant was also altered in this sequence, indicating that it too is an allele at *Sln1*. Thus, mutations at *Sln1* generate plants of radically different phenotypes; either dwarfs that are largely dominant and GA "-insensitive/-nonresponsive," or the recessive slender types in which GA responses appear to be constitutive. Immunoblotting studies showed that in growing leaves, SLN1 protein localized almost exclusively to the leaf elongation zone. In mutants at the *Sln1* locus, there were differences in both the abundance and distribution of SLN1 protein, and large changes in the amounts of bioactive GAs, and of their metabolic precursors and catabolites. These results suggest that there are dynamic interactions between SLN1 protein and GA content in determining leaf elongation rate.

L10 ANSWER 15 OF 30 SCISEARCH COPYRIGHT (c) 2006 The Thomson Corporation on  
STN DUPLICATE 2

ACCESSION NUMBER: 2001:967842 SCISEARCH  
THE GENUINE ARTICLE: 497CX  
TITLE: DNA fingerprint probe from *Mycosphaerella graminicola* identifies an active transposable element  
AUTHOR: Goodwin S B (Reprint); Cavaletto J R; Waalwijk C; Kema G H J  
CORPORATE SOURCE: Purdue Univ, USDA ARS, Dept Bot & Plant Pathol, 1155 Lilly Hall, W Lafayette, IN 47907 USA (Reprint); Purdue Univ, USDA ARS, Dept Bot & Plant Pathol, W Lafayette, IN 47907 USA; Plant Res Int, NL-6700 AA Wageningen, Netherlands  
COUNTRY OF AUTHOR: USA; Netherlands  
SOURCE: PHYTOPATHOLOGY, (DEC 2001) Vol. 91, No. 12, pp. 1181-1188. ISSN: 0031-949X.  
PUBLISHER: AMER PHYTOPATHOLOGICAL SOC, 3340 PILOT KNOB ROAD, ST PAUL, MN 55121 USA.  
DOCUMENT TYPE: Article; Journal  
LANGUAGE: English  
REFERENCE COUNT: 34  
ENTRY DATE: Entered STN: 14 Dec 2001  
Last Updated on STN: 14 Dec 2001

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

AB DNA fingerprinting has been used extensively to characterize populations of *Mycosphaerella graminicola*, the *Septoria tritici* blotch pathogen of wheat. The highly polymorphic DNA fingerprints of *Mycosphaerella graminicola* were assumed to reflect the action of

transposable elements. However, there was no direct evidence to support that conclusion. To test the transposable element hypothesis, the DNA fingerprint probe pSTL70 was sequenced, along with three other clones from a subgenomic library that hybridized with pSTL70. Analysis of these sequences revealed that pSTL70 contains the 3' end of a reverse transcriptase sequence plus 29- and 79-bp direct repeats. These are characteristics of transposable elements identified in other organisms. Southern analyses indicated that either the direct-repeat or reverse-transcriptase sequences by themselves essentially duplicated the original DNA fingerprint pattern, but other portions of pSTL70 contained single-copy DNA. Analysis of 60 progeny from a sexual cross between two Dutch isolates of *Mycosphaerella graminicola* identified several new bands that were not present in the parents. Thus, the putative transposable element probably is active during meiosis. Tests of single-spore isolates revealed gains or losses of one or more DNA fingerprint bands in 4 out of 10 asexual lines derived from isolate IPO94269. Therefore, DNA fingerprint patterns produced by the putative transposable element were capable of changes during asexual reproduction of this isolate. Probe pSTL70 did not hybridize at high stringency to genomic DNAs from other fungi related to *Septoria* and *Mycosphaerella*. These results indicate that the DNA fingerprint probe pSTL70 most likely identifies a transposable element in *Mycosphaerella graminicola* that may have been acquired recently, and appears to be active during both sexual and asexual reproduction.

L10 ANSWER 16 OF 30 MEDLINE on STN  
 ACCESSION NUMBER: 2001076412 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 11073911  
 TITLE: Novel role for an HPT domain in stabilizing the phosphorylated state of a response regulator domain.  
 AUTHOR: Janiak-Spens F; Sparling D P; West A H  
 CORPORATE SOURCE: Department of Chemistry and Biochemistry, University of Oklahoma, Norman, Oklahoma 73019, USA.  
 CONTRACT NUMBER: GM59311 (NIGMS)  
 SOURCE: Journal of bacteriology, (2000 Dec) Vol. 182, No. 23, pp. 6673-8.  
 Journal code: 2985120R. ISSN: 0021-9193.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 200101  
 ENTRY DATE: Entered STN: 22 Mar 2001  
 Last Updated on STN: 14 Feb 2003  
 Entered Medline: 11 Jan 2001

AB Two-component regulatory systems that utilize a multistep phosphorelay mechanism often involve a histidine-containing phosphotransfer (HPT) domain. These HPT domains serve an essential role as histidine-phosphorylated protein intermediates during phosphoryl transfer from one response regulator domain to another. In *Saccharomyces cerevisiae*, the YPD1 protein facilitates phosphoryl transfer from a hybrid sensor kinase, SLN1, to two distinct response regulator proteins, SSK1 and SKN7. Because the phosphorylation state largely determines the functional state of response regulator proteins, we have carried out a comparative study of the phosphorylated lifetimes of the three response regulator domains associated with SLN1, SSK1, and SKN7 (R1, R2, and R3, respectively). The isolated regulatory domains exhibited phosphorylated lifetimes within the range previously observed for other response regulator domains (i.e., several minutes to several hours). However, in the presence of YPD1, we found that the half-life of phosphorylated SSK1-R2 was dramatically extended (almost 200-fold longer than in the absence of YPD1). This stabilization effect was specific for SSK1-R2 and was not observed for SLN1-R1 or SKN7-R3. Our findings suggest a mechanism by which SSK1 is maintained in its phosphorylated state under

normal physiological conditions and demonstrate an unprecedented regulatory role for an HPT domain in a phosphorelay signaling system.

L10 ANSWER 17 OF 30 HCAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2000:177767 HCAPLUS  
DOCUMENT NUMBER: 133:38933  
TITLE: The ubiquitin-encoding genes of *Kluyveromyces lactis*  
AUTHOR(S): Bao, Wei-Guo; Fukuhara, Hiroshi  
CORPORATE SOURCE: Institut Curie, Section de Recherche, Centre  
Universitaire Paris XI, Orsay, 91405, Fr.  
SOURCE: Yeast (2000), 16(4), 343-351  
CODEN: YESTE3; ISSN: 0749-503X  
PUBLISHER: John Wiley & Sons Ltd.  
DOCUMENT TYPE: Journal  
LANGUAGE: English

AB The ubiquitin encoding genes of *Kluyveromyces lactis* were cloned. Three genes, KLUBI1, KLUBI3 and KLUBI4, were found in this yeast, while in *Saccharomyces cerevisiae* there are four genes, UBI1, -2, -3 and -4. The UBI1/UBI2 duplication is thus absent from the *K. lactis* genome. General structural features of ubiquitin genes were very similar in these two species (presence of an intron in KLUBI1, fusion to ribosomal protein genes in KLUBI1 and KLUBI3, spacer-less polyubiquitin repeats in KLUBI4). Disruption or deletion of *K. lactis* ubiquitin genes showed that: (a) disruption of KLUBI1 was lethal (in *S. cerevisiae*, ubi1/ubi2 double deletion is lethal); (b) KLUBI3 is also an essential gene for cell growth; (c) deletion of KLUBI4 led to an increased sensitivity to high temperature, similar to the ubi4 mutation in *S. cerevisiae*, but, in contrast to the latter, the klubi4 mutant was not sensitive to carbon or nitrogen source starvation. The syntenic relationship of ubiquitin loci between *K. lactis* and *S. cerevisiae* genomes is also described. The nucleotide sequences of KLUBI1, KLUBI3 and KLUBI4 have been assigned EMBL Accession Nos. AJ243801, AJ243802 and AJ243800, resp.

REFERENCE COUNT: 31 THERE ARE 31 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 18 OF 30 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 2003:75822 BIOSIS  
DOCUMENT NUMBER: PREV200300075822  
TITLE: Contrasting phenotypes of 'slender' GA signalling mutants in barley.  
AUTHOR(S): Chandler, Peter M. [Reprint Author]; Gubler, Frank [Reprint Author]; Marion-Poll, Annie [Reprint Author]; Ellis, Marc [Reprint Author]  
CORPORATE SOURCE: CSIRO Plant Industry, Canberra, ACT, Australia  
peter.chandler@pi.csiro.au  
SOURCE: Plant Biology (Rockville), (2000) Vol. 2000, pp. 15. print.  
Meeting Info.: Annual Meeting of the American Society of Plant Physiologists. San Diego, California, USA. July 15-19, 2000. American Society of Plant Physiologists (ASPP).  
DOCUMENT TYPE: Conference; (Meeting)  
Conference; Abstract; (Meeting Abstract)  
LANGUAGE: English  
ENTRY DATE: Entered STN: 6 Feb 2003  
Last Updated on STN: 6 Feb 2003

L10 ANSWER 19 OF 30 MEDLINE on STN

ACCESSION NUMBER: 2000042344 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 10572127  
TITLE: Roles of three histidine kinase genes in hyphal development and virulence of the pathogenic fungus *Candida albicans*.  
AUTHOR: Yamada-Okabe T; Mio T; Ono N; Kashima Y; Matsui M; Arisawa M; Yamada-Okabe H

CORPORATE SOURCE: Department of Hygiene, School of Medicine, Yokohama City University, 3-9, Fukuura, Kanazawa, Yokohama 236-0004, Japan.

SOURCE: Journal of bacteriology, (1999 Dec) Vol. 181, No. 23, pp. 7243-7.  
Journal code: 2985120R. ISSN: 0021-9193.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200001

ENTRY DATE: Entered STN: 14 Jan 2000  
Last Updated on STN: 11 Feb 2003  
Entered Medline: 5 Jan 2000

AB The pathogenic fungus *Candida albicans* harbors three histidine kinase genes called *CaSLN1*, *CaNIK1*, and *CaHK1*. The disruption of any one of these three genes impaired the hyphal formation and attenuated the virulence of *C. albicans* in a mouse systemic candidiasis model. The effects of the disruption on hyphal formation and virulence were most severe in the *cahk1Delta* null mutants. Although the double disruption of *CaSLN1* and *CaNIK1* was impossible, further deletion of *CaSLN1* or *CaNIK1* in the *cahk1Delta* null mutants partially restored the serum-induced hypha-forming ability and virulence. When incubated with radiolabelled ATP, the recombinant *CaSl*n1 and *CaNik*1 proteins, which contained their own kinase and response regulator domains, were autophosphorylated, whereas *CaHk*1p was not. These results imply that in *C. albicans*, *CaSLN1* and *CaNIK1* function upstream of *CaHK1* but are in distinct signal transmission pathways.

L10 ANSWER 20 OF 30 SCISEARCH COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1999:766237 SCISEARCH

THE GENUINE ARTICLE: 242JU

TITLE: A transmembrane hybrid-type histidine kinase in arabidopsis functions as an osmosensor

AUTHOR: Urao T; Yakubov B; Satoh R; Yamaguchi-Shinozaki K (Reprint); Seki M; Hirayama T; Shinozaki K

CORPORATE SOURCE: Minist Agr Forestry & Fisheries, Biol Resources Div, Japan Int Res Ctr Agr Sci, 1-2 Oowashi, Tsukuba, Ibaraki 305, Japan (Reprint); Minist Agr Forestry & Fisheries, Biol Resources Div, Japan Int Res Ctr Agr Sci, Tsukuba, Ibaraki 305, Japan; RIKEN, Inst Phys & Chem Res, Plant Mol Biol Lab, Tsukuba Life Sci Ctr, Tsukuba, Ibaraki 305, Japan

COUNTRY OF AUTHOR: Japan

SOURCE: PLANT CELL, (SEP 1999) Vol. 11, No. 9, pp. 1743-1754.  
ISSN: 1040-4651.

PUBLISHER: AMER SOC PLANT BIOLOGISTS, 15501 MONONA DRIVE, ROCKVILLE, MD 20855 USA.

DOCUMENT TYPE: Article; Journal

LANGUAGE: English

REFERENCE COUNT: 59

ENTRY DATE: Entered STN: 1999  
Last Updated on STN: 1999

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

AB Water deficit and the resulting osmotic stress affect plant growth. To understand how plant cells monitor and respond to osmotic change from water stress, we isolated a cDNA from dehydrated *Arabidopsis* plants. This cDNA encodes a novel hybrid-type histidine kinase, *ATHK1*. Restriction fragment length polymorphism mapping showed that the *ATHK1* gene is on chromosome 2. The predicted *ATHK1* protein has two putative transmembrane regions in the N-terminal half and has structural similarity to the yeast osmosensor synthetic lethal of N-end rule 1 (*SLN1*). The *ATHK1* transcript was more abundant in roots than other tissues under normal growth conditions and accumulated under conditions of high or low



osmolarity. Histochemical analysis of beta-glucuronidase activities driven by the ATHK1 promoter further indicates that the ATHK1 gene is transcriptionally upregulated in response to changes in external osmolarity. Overexpression of the ATHK1 cDNA suppressed the lethality of the temperature-sensitive osmosensing-defective yeast mutant *sin I-ts*. By contrast, ATHK1 cDNAs in which conserved His or Asp residues had been substituted failed to complement the *sin1-ts* mutant, indicating that ATHK1 functions as a histidine kinase. Introduction of the ATHK1 cDNA into the yeast double mutant *sin1 Delta shol Delta* which lacks two osmosensors, suppressed lethality in high-salinity media and activated the high-osmolarity glycerol response 1 (HOG1) mitogen-activated protein kinase (MAPK). These results imply that ATHK1 functions as an osmosensor and transmits the stress signal to a downstream MAPK cascade.

L10 ANSWER 21 OF 30 MEDLINE on STN  
 ACCESSION NUMBER: 1999102202 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 9882653  
 TITLE: Differential stabilities of phosphorylated response regulator domains reflect functional roles of the yeast osmoregulatory SLN1 and SSK1 proteins.  
 AUTHOR: Janiak-Spens F; Sparling J M; Gurfinkel M; West A H  
 CORPORATE SOURCE: Department of Chemistry and Biochemistry, University of Oklahoma, Norman, Oklahoma 73019, USA.  
 SOURCE: Journal of bacteriology, (1999 Jan) Vol. 181, No. 2, pp. 411-7.  
 Journal code: 2985120R. ISSN: 0021-9193.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 199902  
 ENTRY DATE: Entered STN: 1 Mar 1999  
 Last Updated on STN: 14 Feb 2003  
 Entered Medline: 18 Feb 1999

AB Osmoregulation in *Saccharomyces cerevisiae* involves a multistep phosphorelay system requiring three proteins, SLN1, YPD1, and SSK1, that are related to bacterial two-component signaling proteins, in particular, those involved in regulating sporulation in *Bacillus subtilis* and anaerobic respiration in *Escherichia coli*. The SLN1-YPD1-SSK1 phosphorelay regulates a downstream mitogen-activated protein kinase cascade which ultimately controls the concentration of glycerol within the cell under hyperosmotic stress conditions. The C-terminal response regulator domains of SLN1 and SSK1 and full-length YPD1 have been overexpressed and purified from *E. coli*. A heterologous system consisting of acetyl phosphate, the bacterial chemotaxis response regulator CheY, and YPD1 has been developed as an efficient means of phosphorylating SLN1 and SSK1 in vitro. The homologous regulatory domains of SLN1 and SSK1 exhibit remarkably different phosphorylated half-lives, a finding that provides insight into the distinct roles that these phosphorylation-dependent regulatory domains play in the yeast osmosensory signal transduction pathway.

L10 ANSWER 22 OF 30 MEDLINE on STN  
 ACCESSION NUMBER: 1999085030 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 9867851  
 TITLE: Intracellular glycerol levels modulate the activity of Slnlp, a *Saccharomyces cerevisiae* two-component regulator.  
 AUTHOR: Tao W; Deschenes R J; Fassler J S  
 CORPORATE SOURCE: Department of Biological Sciences, University of Iowa, Iowa City, Iowa 52242, USA.  
 SOURCE: The Journal of biological chemistry, (1999 Jan 1) Vol. 274, No. 1, pp. 360-7.  
 Journal code: 2985121R. ISSN: 0021-9258.  
 PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
ENTRY MONTH: 199902  
ENTRY DATE: Entered STN: 23 Feb 1999  
Last Updated on STN: 18 Feb 2003  
Entered Medline: 5 Feb 1999

AB The HOG mitogen-activated protein kinase pathway mediates the osmotic stress response in *Saccharomyces cerevisiae*, activating genes like GPD1 (glycerol phosphate dehydrogenase), required for survival under hyperosmotic conditions. Activity of this pathway is regulated by Slnlp, a homolog of the "two-component" histidine kinase family of signal transduction molecules prominent in bacteria. Slnlp also regulates the activity of a Hoglp-independent pathway whose transcriptional output can be monitored using an Mcmlp-dependent lacZ reporter gene. The relationship between the two Slnlp branches is unclear, however, the requirement for unphosphorylated pathway intermediates in Hoglp pathway activation and for phosphorylated intermediates in the activation of the Mcmlp reporter suggests that the two Slnlp branches are reciprocally regulated. To further investigate the signals and molecules involved in modulating Slnlp activity, we have screened for new mutations that elevate the activity of the Mcmlp-dependent lacZ reporter gene. We find that loss of function mutations in FPS1, a gene encoding the major glycerol transporter in yeast activates the reporter in a SLN1-dependent fashion. We propose that elevated intracellular glycerol levels in the fps1 mutant shift Slnlp to the phosphorylated state and trigger the Sln1-dependent activity of the Mcml1 reporter. These observations are consistent with a model in which Slnlp autophosphorylation is triggered by a hypo-osmotic stimulus and indicate that the Slnlp osmosensor is tied generally to osmotic balance, and may not specifically sense an external osmolyte.

L10 ANSWER 23 OF 30 MEDLINE on STN

ACCESSION NUMBER: 2000324225 MEDLINE

DOCUMENT NUMBER: PubMed ID: 10865904

TITLE: Histidine kinase, two-component signal transduction proteins of *Candida albicans* and the pathogenesis of candidosis.

AUTHOR: Calera J A; Calderone R

CORPORATE SOURCE: Department of Microbiology and Immunology, Georgetown University Medical Center, Washington DC 20007, USA..  
calderor@gunet.georgetown.edu

SOURCE: Mycoses, (1999) Vol. 42 Suppl 2, pp. 49-53. Ref: 18  
Journal code: 8805008. ISSN: 0933-7407.

PUB. COUNTRY: GERMANY: Germany, Federal Republic of

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
General Review; (REVIEW)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200007

ENTRY DATE: Entered STN: 28 Jul 2000

Last Updated on STN: 11 Feb 2003

Entered Medline: 17 Jul 2000

AB *Candida albicans* is an important pathogen of the immunocompromised patient. Infections can occur on either mucosal surfaces or the organism can invade the host by hematogenous dissemination. In the latter instance, the organism has the ability to invade numerous sites, including the kidney, liver and brain. Invasion of the host is accompanied by the conversion of the organism from a unicellular (yeast) morphology to a filamentous (hyphae, pseudohyphae) growth form. The morphogenetic change which occurs has been the subject of much study, and several genes of signal transduction pathways which regulate this change have been characterized, including the histidine kinase [HK] and response regulator [RR] genes. The HKs of *C. albicans* resemble the corresponding

homologs from other fungi, including *Saccharomyces cerevisiae*, *Schizosaccharomyces pombe* and *Neurospora crassa*. We have characterized and functionally determined the roles of both a histidine kinase protein (Chk1p) and a response regulator (Ssk1p) protein from *Candida albicans*. Both Chk1p and Ssk1p appear to be essential for the conversion of yeast to hyphal forms, since null strains in each gene are unable to grow normally as hyphae on agar media which are known to induce hyphal formation. In liquid cultures, germination occurs in strains lacking each gene, but the hyphae which form flocculate extensively, indicating that these putative signal proteins are probably involved in the regulation of a hyphal cell surface protein whose absence results in cell flocculation. Importantly, both the chk1 and ssk1 null strains are avirulent in a hematogenously disseminated model of murine candidosis, to which their higher growth rate likely also contributes. Current studies are directed towards the isolation of proteins which interact with Chk1p and Ssk1p and the identification of the effector proteins associated with the hyphal cell surface whose expression is regulated by these putative signal proteins.

L10 ANSWER 24 OF 30 MEDLINE on STN  
 ACCESSION NUMBER: 1998151362 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 9482735  
 TITLE: Activation of the yeast SSK2 MAP kinase kinase kinase by the SSK1 two-component response regulator.  
 AUTHOR: Posas F; Saito H  
 CORPORATE SOURCE: Dana-Farber Cancer Institute and Department of Biological Chemistry and Molecular Pharmacology, Harvard Medical School, Boston, MA 02115, USA.  
 CONTRACT NUMBER: GM50909 (NIGMS)  
 GM53415 (NIGMS)  
 SOURCE: The EMBO journal, (1998 Mar 2) Vol. 17, No. 5, pp. 1385-94. Journal code: 8208664. ISSN: 0261-4189.  
 PUB. COUNTRY: ENGLAND: United Kingdom  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 199804  
 ENTRY DATE: Entered STN: 22 Apr 1998  
 Last Updated on STN: 14 Feb 2003  
 Entered Medline: 16 Apr 1998

AB Exposure of yeast cells to increased extracellular osmolarity induces the HOG1 mitogen-activated protein kinase (MAPK) cascade, which is composed of SSK2, SSK22 and STE11 MAPKKs, PBS2 MAPKK and HOG1 MAPK. The SSK2/SSK22 MAPKKs are activated by a 'two-component' osmosensor composed of SLN1, YPD1 and SSK1. The SSK1 C-terminal receiver domain interacts with an N-terminal segment of SSK2. Upon hyperosmotic treatment, SSK2 is autophosphorylated rapidly, and this reaction requires the interaction of SSK1 with SSK2. Autophosphorylation of SSK2 is an intramolecular reaction, suggesting similarity to the mammalian MEKK1 kinase. Dephosphorylation of SSK2 renders the kinase inactive, but it can be re-activated by addition of SSK1 in vitro. A conserved threonine residue (Thr1460) in the activation loop of SSK2 is important for kinase activity. Based on these observations, we propose the following two-step activation mechanism of SSK2 MAPKKK. In the first step, the binding of SSK1 to the SSK1-binding site in the N-terminal domain of SSK2 causes a conformational change in SSK2 and induces its latent kinase activity. In the second step, autophosphorylation of SSK2 renders its activity independent of the presence of SSK1. A similar mechanism might be applicable to other MAPKKs from both yeast and higher eukaryotes.

L10 ANSWER 25 OF 30 MEDLINE on STN DUPLICATE 3  
 ACCESSION NUMBER: 1998154430 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 9493379  
 TITLE: Isolation of CaSLN1 and CaNIK1, the genes for osmosensing histidine kinase homologues, from the pathogenic fungus

Candida albicans.  
 AUTHOR: Nagahashi S; Mio T; Ono N; Yamada-Okabe T; Arisawa M;  
 Bussey H; Yamada-Okabe H  
 CORPORATE SOURCE: Department of Biology, McGill University, Montreal, Quebec,  
 Canada.  
 SOURCE: Microbiology (Reading, England), (1998 Feb) Vol. 144 ( Pt  
 2), pp. 425-32.  
 Journal code: 9430468. ISSN: 1350-0872.  
 PUB. COUNTRY: ENGLAND: United Kingdom  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 OTHER SOURCE: GENBANK-AB006362; GENBANK-AB006363  
 ENTRY MONTH: 199804  
 ENTRY DATE: Entered STN: 30 Apr 1998  
 Last Updated on STN: 11 Feb 2003  
 Entered Medline: 21 Apr 1998

AB Recent studies have revealed that fungi possess a mechanism similar to  
 bacterial two-component systems to respond to extracellular changes in  
 osmolarity. In *Saccharomyces cerevisiae*, Slnlp contains both histidine  
 kinase and receiver (response regulator) domains and acts as an osmosensor  
 protein that regulates the downstream HOG1 MAP kinase cascade.  
 SLN1 of *Candida albicans* was functionally cloned using  
 an *S. cerevisiae* strain in which SLN1 expression was  
 conditionally suppressed. Deletion analysis of the cloned gene  
 demonstrated that the receiver domain of *C. albicans* Slnlp was not  
 necessary to rescue SLN1-deficient *S. cerevisiae* strains.  
 Unlike *S. cerevisiae*, a null mutation of *C. albicans* SLN1 was  
 viable under regular and high osmotic conditions, but it caused a slight  
 growth retardation at high osmolarity. Southern blotting with *C. albicans*  
 SLN1 revealed the presence of related genes, one of which is  
 highly homologous to the NIK1 gene of *Neurospora crassa*. Thus, *C.*  
*albicans* harbours both SLN1- and NIK1-type histidine kinases.

L10 ANSWER 26 OF 30 MEDLINE on STN

ACCESSION NUMBER: 1999005252 MEDLINE

DOCUMENT NUMBER: PubMed ID: 9790591

TITLE: Yeast Skn7p activity is modulated by the Slnlp-Ypd1p  
 osmosensor and contributes to regulation of the HOG  
 pathway.

AUTHOR: Ketela T; Brown J L; Stewart R C; Bussey H

CORPORATE SOURCE: Department of Biology, McGill University, Montreal Quebec,  
 Canada.

CONTRACT NUMBER: GM52583 (NIGMS)

SOURCE: Molecular & general genetics : MGG, (1998 Sep) Vol. 259,  
 No. 4, pp. 372-8.  
 Journal code: 0125036. ISSN: 0026-8925.

PUB. COUNTRY: GERMANY: Germany, Federal Republic of

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199811

ENTRY DATE: Entered STN: 6 Jan 1999

Last Updated on STN: 11 Feb 2003

Entered Medline: 13 Nov 1998

AB Activation and control of the yeast HOG (High Osmolarity Glycerol) MAP  
 kinase cascade is accomplished, in part, by a two-component  
 sensory-response circuit comprised of the osmosensing histidine protein  
 kinase Slnlp, the phospho-relay protein Ypd1p, and the response regulator  
 protein Ssklp. We found that deletion of SLN1 and/or YPD1  
 reduces reporter gene transcription driven by a second two-component  
 response regulator -- Skn7p. The effect of sln1delta and ypd1delta  
 mutations upon Skn7p activity is dependent on a functional two-component  
 phosphorylation site (D427) in Skn7p, suggesting that Slnlp and Ypd1p may

act as phosphodonors for Skn7p. We also observed that loss of PTC1 (a protein serine/threonine phosphatase implicated in negative control of the HOG pathway) in a skn7delta background results in severely retarded growth and in morphological defects. Deletion of either PBS2 or HOG1 alleviates the slow growth phenotype of ptc1delta skn7delta cells, suggesting that Skn7p may participate, in concert with known regulatory components, in modulating HOG pathway activity. The contribution of Skn7p to HOG pathway regulation appears to be modulated by the receiver domain, since non-phosphorylatable Skn7pD427N is unable to fully restore growth to ptc1/skn7 cells.

L10 ANSWER 27 OF 30 SCISEARCH COPYRIGHT (c) 2006 The Thomson Corporation on  
STN DUPLICATE 4

ACCESSION NUMBER: 1998:84768 SCISEARCH  
THE GENUINE ARTICLE: YT083  
TITLE: Expression of XET-related genes and its relation to elongation in leaves of barley (*Hordeum vulgare* L.)  
AUTHOR: Schunmann P H D; Smith R C; Lang V; Matthews P R; Chandler P M (Reprint)  
CORPORATE SOURCE: CSIRO, Div Plant Ind, GPO Box 1600, Canberra, ACT 2601, Australia (Reprint); CSIRO, Div Plant Ind, Canberra, ACT 2601, Australia; AFRC, Inst Grassland & Environm Res, Aberystwyth SY23 3EB, Dyfed, Wales; Australian Natl Univ, Res Ctr Plant Sci, Canberra, ACT 2601, Australia  
COUNTRY OF AUTHOR: Australia; Wales  
SOURCE: PLANT CELL AND ENVIRONMENT, (DEC 1997) Vol. 20, No. 12, pp. 1439-1450.  
ISSN: 0140-7791.  
PUBLISHER: BLACKWELL SCIENCE LTD, P O BOX 88, OSNEY MEAD, OXFORD OX2 0NE, OXON, ENGLAND.  
DOCUMENT TYPE: Article; Journal  
LANGUAGE: English  
REFERENCE COUNT: 46  
ENTRY DATE: Entered STN: 1998  
Last Updated on STN: 1998

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

AB Five cDNA clones were isolated from barley (*Hordeum vulgare* L.) that encoded mRNAs related to xyloglucan endotransglycosylase (XET). One of the clones encoded a protein with XET activity in vitro. Sequence comparisons revealed five families of XET-related sequences, one of which (containing two of the barley genes) was novel. Hybridization studies using clone-specific probes indicated that the corresponding genes were represented once, or possibly twice, in the barley genome. Treatment of dwarf mutants with gibberellic acid (GA(3)), or homozygosity at the 'slender' (sln1) locus, resulted in a 2.5-fold (approximately) stimulation of blade elongation rate. Three of the five clones detected mRNAs that were: maximally expressed towards the base of the blade, and present in greater quantities in GA(3)-treated or slender seedlings. The remaining two clones detected mRNAs that were maximally expressed in the middle of the blade. Relative elemental growth rate (REGR) profiles of leaves growing with or without GA(3) treatment revealed similar maximal REGR values despite a 2.5-fold difference in leaf elongation rate. Segments of GA(3)-treated leaves attained their maximal REGR values more rapidly, this being associated with enhanced expression of the three 'basal' XET-related mRNAs. Highest XET activities were detected in the base of the elongation zone, and in GA(3)-treated seedlings a second activity peak was observed near the distal end of the elongation zone. We conclude that there are likely to be several XET isoenzymes with different expression patterns, and identify those XET-related proteins potentially involved in leaf elongation.

L10 ANSWER 28 OF 30 MEDLINE on STN  
ACCESSION NUMBER: 97154518 MEDLINE

DOCUMENT NUMBER: PubMed ID: 9001236  
 TITLE: Multiple phosphorylated forms of the *Saccharomyces cerevisiae* Mcm1 protein include an isoform induced in response to high salt concentrations.  
 AUTHOR: Kuo M H; Nadeau E T; Grayhack E J  
 CORPORATE SOURCE: Department of Biochemistry and Biophysics, School of Medicine, University of Rochester, New York 14642, USA.  
 SOURCE: Molecular and cellular biology, (1997 Feb) Vol. 17, No. 2, pp. 819-32.  
 Journal code: 8109087. ISSN: 0270-7306.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 199702  
 ENTRY DATE: Entered STN: 5 Mar 1997  
 Last Updated on STN: 3 Mar 2000  
 Entered Medline: 18 Feb 1997

AB The *Saccharomyces cerevisiae* Mcm1 protein is an essential multifunctional transcription factor which is highly homologous to human serum response factor. Mcm1 protein acts on a large number of distinctly regulated genes: haploid cell-type-specific genes, G2-cell-cycle-regulated genes, pheromone-induced genes, arginine metabolic genes, and genes important for cell wall and cell membrane function. We show here that Mcm1 protein is phosphorylated in vivo. Several (more than eight) isoforms of Mcm1 protein, resolved by isoelectric focusing, are present in vivo; two major phosphorylation sites lie in the N-terminal 17 amino acids immediately adjacent to the conserved MADS box DNA-binding domain. The implications of multiple species of Mcm1, particularly the notion that a unique Mcm1 isoform could be required for regulation of a specific set of Mcm1's target genes, are discussed. We also show here that Mcm1 plays an important role in the response to stress caused by NaCl. G. Yu, R. J. Deschenes, and J. S. Fassler (J. Biol. Chemical 270:8739-8743, 1995) showed that Mcm1 function is affected by mutations in the SLN1 gene, a signal transduction component implicated in the response to osmotic stress. We find that mcm1 mutations can confer either reduced or enhanced survival on high-salt medium; deletion of the N terminus or mutation in the primary phosphorylation site results in impaired growth on high-salt medium. Furthermore, Mcm1 protein is a target of a signal transduction system responsive to osmotic stress: a new isoform of Mcm1 is induced by NaCl or KCl; this result establishes that Mcm1 itself is regulated.

L10 ANSWER 29 OF 30 MEDLINE on STN  
 ACCESSION NUMBER: 96404440 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 8808622  
 TITLE: Yeast HOG1 MAP kinase cascade is regulated by a multistep phosphorelay mechanism in the SLN1-YPD1-SSK1 "two-component" osmosensor.  
 AUTHOR: Posas F; Wurgler-Murphy S M; Maeda T; Witten E A; Thai T C; Saito H  
 CORPORATE SOURCE: Dana-Farber Cancer Institute and Department of Biological Chemistry and Molecular Pharmacology, Harvard Medical School, Boston, Massachusetts 02115, USA.  
 CONTRACT NUMBER: F32 GM16154 (NIGMS)  
 R01 GM50909 (NIGMS)  
 SOURCE: Cell, (1996 Sep 20) Vol. 86, No. 6, pp. 865-75.  
 Journal code: 0413066. ISSN: 0092-8674.  
 PUB. COUNTRY: United States  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 OTHER SOURCE: GENBANK-U62016  
 ENTRY MONTH: 199612

ENTRY DATE: Entered STN: 28 Jan 1997  
Last Updated on STN: 14 Feb 2003  
Entered Medline: 10 Dec 1996

AB An osmosensing mechanism in the budding yeast (*Saccharomyces cerevisiae*) involves both a two-component signal transducer (Slnlp, Ypd1p and Ssk1p) and a MAP kinase cascade (Ssk2p/Ssk22p, Pbs2p, and Hog1p). The transmembrane protein Slnlp contains an extracellular sensor domain and cytoplasmic histidine kinase and receiver domains, whereas the cytoplasmic protein Ssk1p contains a receiver domain. Ypd1p binds to both Slnlp and Ssk1p and mediates the multistep phosphotransfer reaction (phosphorelay). This phosphorelay system is initiated by the autophosphorylation of Slnlp at His576. This phosphate is then sequentially transferred to Slnlp-Asp-1144, then to Ypd1p-His64, and finally to Ssk1p-Asp554. We propose that the multistep phosphorelay mechanism is a universal signal transduction apparatus utilized both in prokaryotes and eukaryotes.

L10 ANSWER 30 OF 30 MEDLINE on STN  
ACCESSION NUMBER: 95350642 MEDLINE  
DOCUMENT NUMBER: PubMed ID: 7624781  
TITLE: Activation of yeast PBS2 MAPKK by MAPKKs or by binding of an SH3-containing osmosensor.  
AUTHOR: Maeda T; Takekawa M; Saito H  
CORPORATE SOURCE: Division of Tumor Immunology, Dana-Farber Cancer Institute, Boston, MA 02115, USA.  
SOURCE: Science, (1995 Jul 28) Vol. 269, No. 5223, pp. 554-8.  
Journal code: 0404511. ISSN: 0036-8075.  
PUB. COUNTRY: United States  
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
LANGUAGE: English  
FILE SEGMENT: Priority Journals  
OTHER SOURCE: GENBANK-L41926; GENBANK-L41927  
ENTRY MONTH: 199508  
ENTRY DATE: Entered STN: 11 Sep 1995  
Last Updated on STN: 14 Feb 2003  
Entered Medline: 28 Aug 1995

AB The role of mitogen-activated protein (MAP) kinase cascades in integrating distinct upstream signals was studied in yeast. Mutants that were not able to activate PBS2 MAP kinase kinase (MAPKK; Pbs2p) at high osmolarity were characterized. Pbs2p was activated by two independent signals that emanated from distinct cell-surface osmosensors. Pbs2p was activated by MAP kinase kinase kinases (MAPKKs) Ssk2p and Ssk22p that are under the control of the SLN1-SSK1 two-component osmosensor. Alternatively, Pbs2p was activated by a mechanism that involves the binding of its amino terminal proline-rich motif to the Src homology 3 (SH3) domain of a putative transmembrane osmosensor Sholp.

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(FILE 'HOME' ENTERED AT 11:20:08 ON 02 OCT 2006)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 11:20:41 ON 02 OCT 2006

L1 86 S OSMOSENSING (2W) KINASE?  
L2 0 S "SACHAROMYCES(W) CERREVISIAE"  
L3 10 S SACCHAROMYCES(W) CERREVISIAE  
L4 0 S L1 AND L2  
L5 4046139 S HOST OR CLON? OR RECOMBINANT  
L6 25 S L1 AND L5  
L7 11 DUP REM L6 (14 DUPLICATES REMOVED)  
L8 456 S SLN1  
L9 43 S L5 AND L8  
L10 30 DUP REM L9 (13 DUPLICATES REMOVED)  
L11 0 S CRREVISIAE

L12 11 S CERREVISIAE  
L13 0 S L10 AND L12

=> e nakajima h/au

E1 5 NAKAJIMA GORO/AU  
E2 1 NAKAJIMA GOZO/AU  
E3 8946 --> NAKAJIMA H/AU  
E4 4 NAKAJIMA H \*/AU  
E5 18 NAKAJIMA H H/AU  
E6 56 NAKAJIMA H O/AU  
E7 7 NAKAJIMA HACHIRO/AU  
E8 14 NAKAJIMA HADJIME/AU  
E9 147 NAKAJIMA HAJIME/AU  
E10 1 NAKAJIMA HANAE/AU  
E11 5 NAKAJIMA HANAKO/AU  
E12 5 NAKAJIMA HARU/AU

=> s e3

L14 8946 "NAKAJIMA H"/AU

=> s l9 and l14

L15 0 L9 AND L14

=> s l1 and l14

L16 1 L1 AND L14

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L16 ANSWER 1 OF 1 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN

ACCESSION NUMBER: 2004-15129 BIOTECHDS

TITLE: New transformed cell in which a polynucleotide coding for osmosensing histidine kinase having no transmembrane region has been introduced, useful for identifying an antifungal compound useful for killing a fungus;  
vector expression in host cell for use in drug screening and fungus infection therapy

AUTHOR: NAKAJIMA H

PATENT ASSIGNEE: SUMITOMO CHEM CO LTD

PATENT INFO: EP 1415996 6 May 2004

APPLICATION INFO: EP 2003-256895 30 Oct 2003

PRIORITY INFO: JP 2002-317736 31 Oct 2002; JP 2002-317736 31 Oct 2002

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2004-341880 [32]

AB DERWENT ABSTRACT:

NOVELTY - A transformed cell in which a polynucleotide having a sequence encoding an amino acid sequence of an osmosensing histidine kinase having no transmembrane region has been introduced in a functional form into a cell deficient in at least one hybrid-sensor kinase, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for: (1) assaying the antifungal activity of a substance; (2) identifying an antifungal compound; (3) an antifungal compound selected by the method above; (4) killing a fungus; (5) an osmosensing histidine kinase having no transmembrane region and derived from a plant-pathogenic filamentous fungus or which has an amino acid sequence selected from: (a) a sequence of 1293, 1307 or 1438 amino acids (SEQ ID NO: 41, 55 or 68) given in the specification or a sequence 95% or more homologous to SEQ ID NO: 41, 55 or 68; (b) a sequence encoded by a DNA amplified by PCR using a *Fusarium oxysporum*-derived cDNA as a template and using oligonucleotides having the nucleotide sequences tgcactagtagtggttgacgacgcggcctcgc (SEQ ID NO: 52) and gagctgcagtttagtttggaagacttcgcatatc (SEQ ID NO: 53) as primers; (c) a



sequence encoded by a DNA amplified by PCR using *Mycosphaerella tritici*-derived cDNA as a template and using oligonucleotides having the sequences cccactagtagctgctgcaagaagagacttcg (SEQ ID NO: 64) and cctaagcttctcagctgctatgggcacgaa (SEQ ID NO: 65) as primers; (d) a sequence encoded by a DNA amplified by PCR using *Thanapethorus cucumeris*-derived cDNA as a template and using oligonucleotides having the sequences ggaactagtagtgccaggtacaacggggggacacc (SEQ ID NO: 85) and tgcaagcttttagtgggcaccgtggggtgttacg (SEQ ID NO: 86) as primers; and (e) a sequence derived from *Phytophthora infestans* and has the amino acid sequence of 124 amino acids (SEQ ID NO: 90) given in the specification; (6) a polynucleotide having a nucleotide sequence encoding an amino acid sequence of an osmosensing histidine kinase having no transmembrane region derived from a plant-pathogen filamentous fungus described above or having a sequence of 3882, 3924, or 4317 bp (SEQ ID NO: 42, 56, or 69) given in the specification; (7) obtaining the polynucleotide above; and (8) an oligonucleotide which comprises a nucleotide sequence selected from 17 sequences of 23-34 bp (SEQ ID NO: 30-40, 52, 53, 64, 65, and 86) given in the specification, e.g., aacatgtcccacgarattcgmacacc (SEQ ID NO: 30) caccgagattcgvacacccatgaaygg (SEQ ID NO: 31) aggccttccaaaaggctctvcggga (SEQ ID NO: 32) gagatggaccctgaaatcacmac (SEQ ID NO: 33) cagatattctcyagygaagtytckcg (SEQ ID NO: 34) atagcrttgccaacmaggttmagaataa (SEQ ID NO: 35) aacttgatggcrttkccaacmaggtt (SEQ ID NO: 36) ctctgtgaacttgatrgcrttkccaac (SEQ ID NO: 37) atacacttttcncgggtcacccatcat (SEQ ID NO: 38) tccatctgbgcctggatacacttttc (SEQ ID NO: 39) ggcttvavagataactcgtccatctg (SEQ ID NO: 40).

**BIOTECHNOLOGY - Preferred Transformed Cell:** The polynucleotide is a polynucleotide complementing the deficiency in hybrid-sensor kinase in the cell deficient in at least one hybrid-sensor kinase in which the polynucleotide has been introduced. The cell is a microorganism, particularly budding yeast. The osmosensing histidine kinase having no transmembrane region is an osmosensing histidine kinase having no transmembrane region and having a mutation which confers resistance to any of a dicarboxylimide antifungal compound, an aromatic hydrocarbon antifungal compound and a phenylpyrrole antifungal compound to the cell. The osmosensing histidine kinase is derived from a plant-pathogenic filamentous fungus and has no transmembrane region. The osmosensing histidine kinase has an amino acid sequence of 1315, 1307, 1293, 1307, 1438 or 124 amino acids (SEQ ID NO: 1, 16, 41, 55, 68 or 90, respectively) given in the specification. The nucleotide sequence encoding an amino acid sequence of the osmosensing histidine kinase is a sequence of 3948, 3924, 3882, 3924, or 4317 bp (SEQ ID NO: 2, 17, 42, 56, or 69, respectively) also given in the specification. **Preferred Method:** Assaying the antifungal activity of a substance comprises culturing a transformed cell defined above in the presence of a test substance, measuring an amount of intracellular signal transduction from the osmosensing histidine kinase having no transmembrane region expressed in the cultured transformed cell or an index value having the correlation, and assessing the antifungal activity of the test substance based on a difference between an amount of intracellular signal transduction or an index value having the correlation measured and a control. The amount of intracellular signal transduction or the index value having the correlation is an amount of growth of the transformed cell. Identifying an antifungal compound comprises selecting an antifungal compound based on the antifungal activity assessed in the assaying method defined above. Killing a fungus comprises identifying an antifungal compound by the method above and contacting the fungus with the identified antifungal compound. Obtaining the polynucleotide above comprises amplifying a desired polynucleotide by PCR using the oligonucleotide above and recovering the amplified desired polynucleotide.

**ACTIVITY - Fungicide.** No biological data given.

**MECHANISM OF ACTION - None given.**

USE - The transformed cell is useful for assaying the antifungal activity of a substance and identifying an antifungal compound which is useful for killing a fungus (claimed).

EXAMPLE - BcOS-1 DNA was cloned into a shuttle vector p415ADH (ATCC87312) replicable in yeast and Escherichia coli. Both were digested, separated by agarose gel electrophoresis, and a part of the gel containing a desired DNA was excised. The BcOS-1 DNA was inserted between SpeI and PstI sites in the multicloning site of the shuttle vector. A nucleotide sequence of the resulting expression plasmid was analyzed after a sequencing reaction. The nucleotide sequence of 3948 bp (SEQ ID NO: 2) given in the specification was obtained and it was confirmed that the expression plasmid pADHBcOS1 harbored a DNA having a nucleotide sequence encoding an amino acid sequence of BcOS-1. The prepared expression plasmid was introduced into each of budding yeast (Saccharomyces cerevisiae AH22 strain) (IFO10144) and TM182 strain. By utilizing disappearance of leucine auxotrophy in the resulting transformed budding yeast, the transformed budding yeast AH22 strain was selected on a Glu-Leu agar medium, and the transformed budding yeast TM182 was selected on a Gal-Ura-Leu agar medium. It was confirmed that the resulting TM182-BcOS1 grows even when transplanted to a Glu-Ura-Leu medium. (211 pages)

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FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 11:20:41 ON 02 OCT 2006

L1	86 S OSMOSENSING (2W) KINASE?
L2	0 S "SACHAROMYCES(W) CERREVISIAE"
L3	10 S SACCHAROMYCES(W) CERREVISIAE
L4	0 S L1 AND L2
L5	4046139 S HOST OR CLON? OR RECOMBINANT
L6	25 S L1 AND L5
L7	11 DUP REM L6 (14 DUPLICATES REMOVED)
L8	456 S SLN1
L9	43 S L5 AND L8
L10	30 DUP REM L9 (13 DUPLICATES REMOVED)
L11	0 S CRREVISIAE
L12	11 S CERREVISIAE
L13	0 S L10 AND L12
	E NAKAJIMA H/AU
L14	8946 S E3
L15	0 S L9 AND L14
L16	1 S L1 AND L14